THE WANDERING SCHOLAR IN THE BRITISH COMMONWEALTH

A PASSAGE in the Queen's Speech at the opening of the new Parliament last autumn promised legislation to implement recommendations of the Commonwealth Education Conference, and in seconding the address, Mr. E. Gardner not only stressed the importance of seeing that to-morrow's leaders of the Commonwealth have the best and widest education we can give, but also urged the need of a two-way exchange of ideas and learning. He suggested that we should not only welcome Commonwealth students in our universities, our Inns of Court and technical colleges, but should also send our own students to the new universities of Africa and Malaya. In the same debate, Mr. Anthony Head urged much the same from a different point of view, suggesting indeed that it is vital for the West as a whole to overhaul its educational approach to the underdeveloped countries.

Two Bills have already been introduced during the new session which deal with the general problem. The first of these, the Marshall Scholarships Bill, which received its second reading in the House of Commons on November 6 and in the House of Lords on November 26, is essentially a Bill which doubles the number of Marshall Scholarships coming to Great Britain. In moving the second reading, the Joint Under-Secretary of State for Foreign Affairs, Mr. R. Allan, said that during the six years the scheme has been in force, 72 American scholars, including 20 women, had been selected from 37 different colleges and universities in the United States, of whom nine had come from Harvard, six from Stanford, California, and six from Princeton, New Jersey. Twenty-five of the scholars went to Oxford, twenty to Cambridge and fifteen to London, the remainder being spread over eight other British universities; the Marshall Aid Commission would like to see the scholars distributed among more British universities but felt obliged to offer them the university of their choice if vacancies existed there. All in all, the scholarships had achieved the success that had been hoped for, and many suggestions had been made to increase the number of scholars.

The Bill received a warm welcome in both Houses. In the Commons, Mr. P. Noel-Baker associated himself with the tribute paid to the work of the selection commissions in the United States and hoped that these commissions would continue to choose their scholars by regions. Selection simply on academic standards might lead to far too many of the scholarships going to the great universities of the eastern coast of the United States. Mr. P. Goodhart thought that at least one scholarship every year might with advantage go to a graduate of one of the American Service academies, or to the Virginia Military Institute, where General Marshall himself had studied. Mr. M. Stewart referred to the value of such interchanges in interpreting one country to another, and for that reason, he said, it is desirable that the Marshall scholars should be drawn, so far as possible, from every aspect of American life and that they should be given a very wide experience of Britain. It is right that we should respect the scholar's right of choice, but all the more important that we should build up both the performance and the prestige of the newer universities.

Other speakers were just as strongly in support of the scheme, but were anxious to improve it along these lines. Mr. M. MacPherson, for example, wondered whether it would be better to select one student from each State over a period of two years, and suggested that instead of universities in Britain as a group offering places to the twenty-four Marshall scholars, each university should be asked individually to offer a place in a particular subject which it wished to suggest. This might assist better use of some of the special facilities of particular universities, and also correct the concentration on too few subjects. At present students are mainly in social and political science, physics, chemistry and mathematics; of sixty students in the first five years, only two were taking applied science.

While these specific suggestions found little support in the debate, it was generally recognized as desirable to spread the students around so far as possible; and referring to the small number of Marshall scholars studying science, Mr. Allan, replying on the debate, pointed out that the National Science Foundation provides thirty-eight fellowships for Americans to study science. He also promised to put the points raised personally before the chairman of the Commission. In the House of Lords, the Minister of State for Colonial Affairs, the Earl of Perth, who moved the second reading, said that if experience showed that a greater number of scholars than twenty-four was desirable, the Bill would permit such an increase by Order in Council. Indeed, here even more than in the House of Commons, the debate stressed the value of the wandering scholar and the contribution to world understanding which the free intercourse of scholars could offer.

The second Bill, the Commonwealth Scholarships Bill, which received its second reading in the House of Commons on November 25, is the first of the measures promised in the Queen's Speech to give effect to some of the recommendations of the Commonwealth Education Conference. Specifically, it gives effect to the Commonwealth Scholarship and Fellowship scheme described in the White Paper and is intended to introduce a greater measure of academic mobility throughout the Commonwealth at a time when the men and women concerned are at the formative period of their professional careers. As Mr. C. J. M. Alport, Minister of State, Commonwealth

Relations Office, said in moving the second reading, it should provide a continuous exchange of ideas and knowledge by giving them access to the rich and varied intellectual resources and technical experience of the different Commonwealth countries; and he hoped it would go far to create in a very real and literal sense a Commonwealth of the mind, which, applied to the social, economic and political problems of our time, might provide answers to questions which those in the world of public affairs sometimes found so elusive.

The Scholarship and Fellowship Plan is outlined in the report of the Commonwealth Education Conference and the proposed arrangements for the administration of the scheme in the United Kingdom are described in a subsequent White Paper*. The plan is additional to, and distinct from, any other plan already in operation and is based on mutual co-operation and the sharing of educational experience among all countries of the Commonwealth. It is sufficiently flexible to take account of the diverse and changing needs of the countries of the Commonwealth and, while Commonwealth-wide, is operated on the basis of a series of bilateral arrangements to allow for the necessary flexibility. The awards are intended to be designed to recognize and promote the highest standards of intellectual achievement.

If the plan is to achieve its purpose, we must secure the widest possible variety of cultural exchange between all parts of the Commonwealth and so facilitate the development of a multilateral trade in ideas. The Commonwealth Scholarships will represent a system of awards to men and women from all Commonwealth countries who are of high intellectual promise and may also be expected to make a significant contribution to life in their own countries on their return from overseas. In the main, these awards will be made for a period of post-graduate study or research, to men and women capable of reaching the highest standard of intellectual achievement, and the ages of those considered for awards may vary according to the circumstances of particular countries. A limited number of Commonwealth Visiting Fellowships will also be made to senior scholars of established reputation and achievement.

The main emphasis will be on awards in the academic field; but the possibility is not excluded of some awards to other persons who play important parts in the life of their community. Because the awards will be made to persons of high intellectual attainment and held in institutions of higher learning, such institutions and members of the academic community who work in them should play an important part in selecting and placing the scholars and fellows. In the first place, the United Kingdom Government has undertaken to be responsible for the award of a total of 500 scholarships and fellowships to be held at any one time in United Kingdom universities, colleges of technology or other appropriate institutions, and Mr. C. J. M. Alport hoped that the first 250 of these would be available to be taken up at the beginning of the academic year in the autumn of 1960. The Bill creates a Commonwealth Scholarship Commission which will be solely responsible for selecting and placing the Scholars and Fellows. The Commission, of not less than ten or more than fifteen members, at least four of whom will be leading figures in the academic world, including a chairman designated jointly by the Secretary of State for Commonwealth Relations and the Secretary of the Colonies, in accordance with whose directions the Commission will act, will be assisted by a panel of forty advisers consisting of distinguished United Kingdom scholars and scientists. The Association of Universities of the British Commonwealth will provide the secretariat of the Commission, and the British Council will be responsible for paying the cost of passages, fees and allowances and for all arrangements for reception and welfare. When the plan is in full operation, it is expected that the 500 scholarships will involve annual expenditure of about £500,000, with some additional expenditure to supplement awards offered to United Kingdom citizens by other countries of the Commonwealth.

Mr. Alport added that each Commonwealth country is setting up a similar body with broadly the same duties as the Commission in Britain. That Commission will be solely responsible for selecting individuals for scholarships here or recommending candidates for scholarships overseas. He estimated the financial cost at £2,300,000 during the first five years, and urged that in future a period of service or study in the Commonwealth should be regarded not as a handicap, but as an essential qualification for a successful career in the professions, industry or technology in the United Kingdom. He also emphasized the importance of ensuring that students coming to Britain had an opportunity of entering fully into the ordinary life of the country, especially during vacations, and said that special attention would be paid to this by the British Council.

The Bill was warmly welcomed on all sides of both Houses of Parliament, and the debates were marked by much constructive criticism. Mr. H. A. Marquand stressed its value for the exchange of ideas and the exploration of ideas to free the human spirit. The importance of a wide distribution of students using the universities of the United Kingdom was stressed, and the Under-Secretary of State for Commonwealth Relations, Mr. R. Thompson, assured the House that it is intended that the Scholars should go to colleges of technology and not solely to universities. In the House of Lords debate on December 8, the Earl of Home, replying for the Government, anticipated that ultimately some 50 per cent of the Scholars might be studying technical subjects.

The need for special care in the reception of the Scholars was generally appreciated, and in a constructive speech Mr. Geoffrey Lloyd referred to the need to strengthen administrative arrangements in Britain to further the scheme. He suggested also that educational attachés might be useful and that

* Commonwealth Relations Office: Colonial Office, Commonwealth Scholarship and Fellowship Plan: Proposed Arrangements for the Administration of the Plan in the United Kingdom. Pp. 5. (Cmnd. 894.) (London: H.M. Stationery Office, 1959.) 6d. net.

we should consider their appointment to the offices of British High Commissioners and especially those in the younger Commonwealth countries. We must seek also to create an entirely new climate of opinion upon the whole question of Commonwealth education, and Mr. P. Goodhart regarded the Bill as a challenge to the people of Great Britain. The Scholarships would be a direct benefit both to Britain and to the Commonwealth as a whole only if the Scholars and Fellows were not allowed to feel lonely, unwanted and without friends.

In replying on this debate, Mr. R. Thompson remarked that the Government would make money available to remove some of the financial obstacles to the recruitment of teachers for the overseas service, and also said that the United Kingdom Government is planning that an additional 500 places in teacher-training establishments of all kinds should be made available for Commonwealth students over the next five years. There was some comment in both Houses on the absence of the Union of South Africa, which has so far not decided to participate in the scheme. Canada is to provide 500 scholarships and fellowships; Australia, 100; India, 100; Pakistan, 30; New Zealand, 25; the Federation of Malaya, 12; Ghana, 10; the Federation of Rhodesia and Nyasaland, 10; Ceylon, 6; and East Africa, 4. Mr. Marquand raised the point particularly in respect of the High Commission Territories in Africa, and thought that now that the universities of South Africa are closed to students from such territories their needs should be specially considered. Dr. H. King referred specifically to the provision by the students of the University of Southampton of a scholarship deliberately aimed at bringing to England some of the students whom the South African Government had denied proper opportunities of education.

At the Committee stage of the Bill in the House of Commons on December 3, comparatively few fresh points were raised: the importance of wide distribution of the Scholars, while giving due consideration to individual preferences, was again stressed; and, for the Government, Mr. C. J. M. Alport emphasized the importance of spreading these Fellows and Scholars as widely as possible over the academic life of the United Kingdom, both geographically and from the point of view of the subjects they came to study. The question of South Africa was considered rather more fully in the House of Lords debate on December 8. The Earl of Home, in introducing the Bill, added little to what had been said in the House of Commons, but stressed the need to ensure that Scholars or Fellows did not suffer in their careers through going abroad for a couple of years in their training. The question of South Africa, however, was raised in forthright manner by the Earl of Lucan. While we would welcome South African participation in the scheme and also South African students in Britain, he did not think it would be consistent with the conception of the Commonwealth on which the Education Conference was based to agree to British students going to South African universities which had been forced to abandon their old liberal traditions and conform to the present racial policy of the Union.

The Commonwealth, it is maintained in the report of the Conference, is a new experiment in human relationships, founded on a belief in the worth and dignity of the individual and a recognition of the value of freedom and co-operative action. Lord Brand, however, argued that it would not be an offence against human dignity to permit a Scholar from Britain to go to the self-governing University of the Witwatersrand, although the University's protests against the policy of the Union Government had proved ineffective. The Earl of Home, however, pointed out that if South Africa decided to come in and offered scholarships in the South African universities, the decision to apply for such scholarships would rest with the students themselves.

Nevertheless, as a side-issue of the debates, the policy of the South African Government towards the universities stood out in the same stark isolation to that of the remaining Commonwealth countries as it had done at the Conference on "Freedom and Responsibility: the Role of the Scholar in Society" arranged by the Committee on Science and Freedom, of which the report has just been published*. This was apparent in Dr. J. W. Cook's introductory paper on university traditions with particular reference to the British Commonwealth, in which he emphasized the importance of academic autonomy and academic freedom, of avoiding any steps which would hinder the free exchange of information, maintaining high academic standards and avoiding becoming involved in political controversy; and also in Dr. F. G. Malherbe's paper on university problems in a multicultural society, which showed factually how in South Africa the Government policy is depriving millions of non-whites of any chance of a university education. Dr. Malherbe pointed out that so far the academic freedom and autonomy of the existing universities continues to stand high; but a note on dismissals of staff at Fort Hare University College circulated with the report raises grave doubts as to how far this can continue.

The debates in Parliament on both these Bills show that Parliament generally is well aware of their importance and of the value of the wandering scholar in the life of the Commonwealth. It is realized, too, that such interchange can only be fully effective if supported by widespread co-operation both on the part of employers to see that the returning Scholars and Fellows are not affected adversely in their careers and equally on the part of the general public in the receiving countries to ensure that, especially in their leisure hours, they are absorbed freely and warmly into the life of the community. Perhaps too little was said as to the value of interchange, not merely in both directions between the United Kingdom and other Commonwealth countries, but also between the Commonwealth countries themselves. Nevertheless, neither the White Paper itself,

* Committee on Science and Freedom. Bulletin No. 13: The Scholar and Society—Report on a Study Group held in Tunis, April 1959. Pp. 144. (Manchester: Committee on Science and Freedom, 818 Wilmslow Road, 1959.)

the Conference report nor the debates in Parliament can leave anyone in doubt as to the value of these imaginative schemes in promoting understanding and goodwill and increasing the supply of trained people, or in developing in responsible men and women the qualities of judgment and wisdom, insight and sympathy, which the advance of civilization increasingly demands.

RIVER STUDIES IN NIGERIA

River Studies and Recommendations on Improvement of Niger and Benue

By Nedeco, Netherlands Engineering Consultants, The Hague. Pp. ix+1000. (Amsterdam: North-Holland Publishing Company, 1959.) 140*s*.

ACCESS from the sea to the Benin ports of Burutu, Sapele and Warri in Nigeria has given much concern ever since about 1930. These ports constitute the link with the inland navigation of the extensive Niger and Benue river system, which provides a waterway of more than a thousand miles into the continent of Africa. At one time ships drawing up to 20 ft. could cross the bar of the Forcados estuary but in more recent years this estuary, along with the Escravos estuary, has only been able to accommodate ships drawing 12–14 ft. A continual decline in the draught available led the Nigerian Government to call in the Netherlands engineering consultant firm of Nedeco to undertake the Western Niger Delta investigation of 1952–54.

Soon after the Western Delta investigation was started preparations were made for a study of the possibility of improving the shipping conditions on the Niger and Benue Rivers. This eventually led the Nigerian Government to request from Nedeco a full-scale investigation of the Niger and Benue river system with the object of determining how the shipping conditions on these rivers could be effectively improved. The consultants were charged with discovering the characteristic nature and behaviour of the component parts of the river system; the composition by means of aerial and other surveys of a series of river maps covering the rivers downstream from Baro and Guara; of ascertaining the régime of the rivers and the vagaries of channel configuration and depths; and inquiring into the feasibility of possible future projects.

The work was commenced in 1954 and Nedeco has now issued a monumental tome of more than a thousand pages illustrated by several hundred diagrams and maps giving the results of the investigations of a team of twelve expert engineers who were stationed in Nigeria for three years. At first glance the report might be thought to be of interest only to those concerned with the practical problems of river navigation, but closer examination reveals that it will be of great value to many other experts. For the consultants found themselves obliged to consider matters which were not directly concerned with the technical problems of river regulation and river management. Since the Niger and Benue systems drain the greater part of Nigeria, the report effectively becomes an up-to-date account of the geography of Nigeria in which the physiography, geology, climatology and hydrology are all systematically considered as a background to the economic traffic on the rivers. It is these that are largely the life-giving arteries of the country. Of the various sections the hydrological naturally receives the most detailed treatment as this was, of course, a prime charge on the consultants. The report concludes with an account of the present problems of navigation, defects of navigability and recommendations for their improvement.

Because the report is so comprehensive it will be found of interest to geographers, geologists, geomorphologists, climatologists, hydrologists and hydraulic engineers: it brings together a great mass of new information and incorporates many new ideas. There can be few of these experts who will not find something of immediate interest in the report's thousand pages. The whole publication is beautifully produced and illustrated, and Nedeco is indeed to be congratulated on the appearance of such an impressive report.

The Western Niger Delta investigation during 1952–54 dealt with access to the ports of Burutu, Sapele and Warri and therefore only that part of the Niger Delta was studied and surveyed which lies west of the Bomadi Creek. Having undertaken the main Niger–Benue investigation it was only natural that the Government began to think in terms of a hydrological survey and investigation of the remaining part of the Delta. Accordingly, an agreement was signed in March 1958 between the representative of the Government of the Federation of Nigeria and of Nedeco for an Eastern Niger Delta investigation. Field-work for this is now under way, and the final report is expected towards the end of 1960.

W. G. V. Balchin

SOLUTIONS OF ELECTROLYTES

The Physical Chemistry of Electrolytic Solutions

By Prof. Herbert S. Harned and Prof. Benton B. Owen. Third edition. (American Chemical Society Monograph Series, No. 137.) Pp. xxxii+803. (New York: Reinhold Publishing Corporation; London: Chapman and Hall, Ltd., 1958.) 160*s*.

Electrolyte Solutions

The Measurement and Interpretation of Conductance, Chemical Potential and Diffusion in Solutions of Simple Electrolytes. By Prof. R. A. Robinson and Prof. R. H. Stokes. Second edition. Pp. xv+559. (London: Butterworths Scientific Publications; New York: Academic Press, Inc., 1959.) 65*s*.

The Structure of Electrolytic Solutions

Edited by Walter J. Hamer. (Sponsored by the Electrochemical Society, Inc., New York, N.Y., and the National Science Foundation, Washington, D.C.) (Electrochemical Society Series.) Pp. xii+441. (New York: John Wiley and Sons, Inc.; London: Chapman and Hall, Ltd., 1959.) 148*s*. net.

OF the three volumes under review, Harned and Owen's "Physical Chemistry of Electrolytic Solutions" and Robinson and Stokes's "Electrolyte Solutions" are new editions of well-known works that first appeared in 1943 and 1955, respectively. "The Structure of Electrolytic Solutions" is a collection of papers given at an Electrochemical Society symposium in Washington in 1957.

Unlike the second edition, which was largely a reprint of the first, the new edition by Harned and Owen incorporates some major revision and has been considerably extended. A section on irreversible

thermodynamics has been added, and recent theoretical work on inter-ionic forces and their effects on both equilibrium and transport properties is included. Diffusion of electrolytes is fully discussed. The title "The Physical Chemistry of Electrolytic Solutions" is a pardonable exaggeration; many physico-chemical properties of such solutions are ignored. For example, there is little or nothing on reaction kinetics or spectroscopic properties and, although the adsorption of ions at the air/water interface is discussed, there is no mention of electrocapillarity or electrode reactions. The book is primarily concerned with the thermodynamics and transport properties of solutions and the central theme is the relation of such properties to ionic interaction. Broader and less tractable questions, such as standard free-energies and entropies of hydration, or the limiting mobilities of ions and their relation to ion-solvent interaction, receive less attention. Experimental data are fully discussed and tabulated, but experimental methods are only indicated in outline. In its own field this is a far more comprehensive monograph than any other, and much of its contents cannot be found elsewhere except in the original papers. The treatment is nearly always clear and precise, and the amount of detail given is very nicely judged, being neither too much nor too little. The earlier editions have become accepted as a standard and indispensable work of reference, and I do not doubt that the third edition will continue this tradition.

"Electrolytic Solutions", by Robinson and Stokes, is about half as long as Harned and Owen's book, and costs about two-fifths as much. It may be regarded as an advanced text-book (it certainly serves this purpose well), though in some respects it is rather more specialized in character than a text-book is expected to be. It covers a considerable part of Harned and Owen's territory, but generally in much less detail—with the important exception of experimental methods, which are here described fairly fully. In addition, more attention is paid to the interpretation of the properties of ions in terms of their interaction with the solvent. Profs. R. A. Robinson and R. H. Stokes have deliberately restricted themselves to the discussion of properties that seem to them to be fundamental, and as the sub-title indicates, the main topics are the measurement and interpretation of conductance (including transport numbers), chemical potential and diffusion. Thus, the book is by no means a text-book of general electrochemistry, and even in the narrower field of thermodynamics of electrolyte solutions, it does not deal with heats of dilution. But within their self-imposed limits, the authors have given a stimulating and well-balanced account of the subject, and their book can be recommended without hesitation. The first edition contained extensive tables of experimental results; these have now been enlarged and brought up to date and form a most useful collection of data. Elsewhere some minor changes have been made; for example, recent work on conductance in non-aqueous solvents has been incorporated.

The title "The Structure of Electrolytic Solutions" provides a wide umbrella, under which some twenty-seven papers have found shelter. They cover a big range, both in subject and in character. Some are essentially reviews of work with which their authors have been associated, others give new experimental results and a few are theoretical contributions. A few titles chosen at random include "Proton Transfer in Aqueous Solution", "Ionic Vibration Potentials", "Soret Coefficients", "Conductance and *p*H in Methanol–Water Mixtures", "Complex Ions in Molten Salts". It is a useful collection of papers and gives the reader a very fair picture of current trends in this field.

All three books are well produced and none suffers from an undue number of misprints or obvious errors. The price of Harned and Owen is formidable, but it is good value for money. J. N. AGAR

TROPICAL MARINE MOLLUSCS FROM AMERICA

Sea Shells of Tropical West America

Marine Mollusks from Lower California to Colombia. By Prof. A. Myra Keen. Pp. viii+624+10 plates. (Stanford, Calif.: Stanford University Press; London: Oxford University Press, 1958.) 100*s*. net.

THE wealth of illustrated handbooks on American sea-shells available for coasts of the United States (and less expectedly for other areas) is in sad contrast to the dearth of European manuals. Though several faunal series provide identifications, by means of keys and descriptions, of animals in the North and Baltic Seas, no fully illustrated and reasonably comprehensive handbook on the sea-shells of any European coast has appeared for twenty-five years. For a British book, it is necessary to go back twice that time. Let us hope that the recently mooted European Malacological Union may stimulate the production of a work to challenge the best American models.

Among these, Prof. Keen's book stands high. Within well-chosen limits (excluding only shell-less forms, squid and cuttlefish, minute gastropods, stragglers and deep-water forms) she deals with even the rarest and the most dubious Panamic species, and illustrates all but a few. The coloured plates are decorative, while much of the value of the book lies in nearly seventeen hundred monochrome figures, of generally high quality. These are grouped on uniform pale grey backgrounds into half-tone plates, set in the text so that the illustration of a species occurs on the same pair of facing pages as the concise diagnostic notes. A few explanations of the figures, relegated to an appendix, would be helpful in the text. Though relatively small in bulk, the general matter is well conceived, touching on the ecology, affinities and exploration of the Panamic malacofauna, referring to general texts, and discussing the higher taxa. Ecological information on individual species is inevitably scanty, but the book itself will stimulate the observations by which the deficiency will be rectified.

In such a work, intended mainly for the knowledgeable amateur and the general zoologist, the precise systematic arrangement adopted is of secondary importance; but broader genera might have been adopted in a few highly split groups. Also, subgeneric names would tend less to emphasize difference at the expense of likeness, if merely listed at the head of the contained species, instead of being combined in every species name. Indeed, a slight over-emphasis on nomenclature is the most serious charge that can be levelled against Prof. Keen—whose identification of taxonomy with nomenclature will

be ill-received by taxonomists and etymologists alike. That misleading technical term 'type' occurs repeatedly in the addenda though not in the glossary, but will not entrap those who read the suggested works on taxonomic practice. However, the discussion on the name *Mitra sulcata* is comprehensible only, and with difficulty, in the light of a new provision on the status of junior primary homonyms, which many will be surprised to find in the forthcoming edition of the International Rules of Zoological Nomenclature.

The addenda, in fact, are intended not for the general reader but for the systematic malacologist; who should acquire three additional pages, issued separately. They reinforce a bibliography of nearly six hundred titles, with every original reference to an included species, and a large number of original figures reproduced from inaccessible works, to make the handbook invaluable, especially to the isolated worker. Relatively few additional titles would be needed to provide references to the genera also, and publications of one author in one year could be distinguished in the text by means of the letters used in the bibliography. Perhaps these minor improvements may be incorporated in the successive editions which will certainly and deservedly be demanded.

I. C. J. Galbraith

BACTERIOPHAGES

Bacteriophages

By Mark H. Adams. With chapters by E. S. Anderson, J. S. Gots, F. Jacob and E.-L. Wollman. Pp. xviii+592. (New York: Interscience Publishers, Inc.; London: Interscience Publishers, Ltd., 1959.) 115*s*.

BACTERIOPHAGES are viruses made up of genetically active deoxyribonucleic acid enclosed within a protein shell, which seem to have no enzymes of their own, with the exception of some forming part of the specialized apparatus, the phage tail, concerned with their attachment to and penetration of the host. Yet the entry of the phage nucleic acid into a susceptible bacterium results in a complete change-over in the metabolism of the host, with synthesis of new enzymes, proteins and nucleic acids.

The advantages of such a system for the investigation of the mechanisms by which specific biosyntheses are controlled and performed are evident. Furthermore, phages, from the technical point of view, are much more convenient to work with than any of the other known classes of virus. It is therefore not surprising that in recent years the phage-infected bacterium has become a favourite experimental material for fundamental research in cell physiology, biochemistry and, perhaps most important, in physiological genetics, and that our knowledge of phages has increased enormously. Phage has been used as experimental material in such a variety of fields that the reports are scattered to an exceptional degree. The first text-book on phage to appear in English for many years has therefore been eagerly awaited. The work in question was begun some years ago by Dr. Mark Adams, of New York University College of Medicine, well known for his own researches on phage, particularly in the neglected field of their taxonomy, and for the course which he taught at Cold Spring Harbor for many summers; a course which served as an introduction to phage for many biologists and workers from other fields who have since contributed largely to our increasing knowledge of this subject.

At the time of Mark Adams's untimely death in 1956, his manuscript was unfinished. It has been completed by the addition of four chapters, planned but not drafted by Adams: on "Chemical Interference with Phage Growth", by J. S. Gots; on "Lysogeny" and on "Colicins", by F. Jacob and E.-L. Wollman; and on "Use of Phages in Epidemiological Studies", by E. S. Anderson. The eighteen chapters finished or partly written by Adams have been revised, brought up to date and completed by a panel of distinguished workers in phage research, the whole work being under the general editorship of A. D. Hershey.

The book as thus completed forms an excellent and reasonably detailed account of our knowledge of phage. The style and presentation are satisfactory, and even those without previous knowledge of the subject should have no difficulty in following the argument. Indeed, it may perhaps be said that at some points simplification has been carried too far; for example, the equation derived by Visconti and Delbrück for the frequency of a recombinant class among the progeny phage released after infection of bacteria by a mixture of phages of differing genotypes is given, but not its derivation. However, the qualitative treatment given will probably suffice for most readers. Those requiring more detail will find themselves well served by the extensive bibliography (some 900 references); it is to be regretted that not all the citations include the titles of papers. A glossary of terms used in the literature is also included in the volume.

Despite the rapid progress of research on phage, most experiments even now make use of a rather limited number of more or less standard techniques. One of Mark Adams's most useful services to the phage world was the publication in 1950 of a review entitled "Methods of Study of Bacterial Viruses", in which all these indispensable experiments and techniques were described and discussed. This most valuable compendium has been included as an appendix, with such additions and amendments as were needed to bring it up to date.

All the principal aspects of phage are dealt with, and it is satisfactory to be able to record that the earlier literature, mainly European, has not been neglected. The use of phages in epidemiology is the only application of phage described; their use in the treatment of bacterial infections, to which so much attention was given in the years immediately following the discovery of phage, is scarcely mentioned, an omission perhaps justified by the disappointing or equivocal results obtained, both in experimental and natural infections. In general, it may be said that the selection of results from the extensive literature has been admirably made, for while nearly every report of general importance is discussed or mentioned, the very large number of papers of only ephemeral or specialized interest are not referred to.

This work can therefore be warmly recommended (and indeed its high price seems to me its only major fault). It will certainly become, and long remain, the standard source of information on phage. It forms a worthy memorial to its principal author, both as scientist and teacher.

B. Stocker

ELECTRONIC SECTOR-SCANNING ASDIC: AN IMPROVED FISH-LOCATOR AND NAVIGATIONAL INSTRUMENT

By Prof. D. G. TUCKER and Dr. V. G. WELSBY

Electrical Engineering Department, University of Birmingham

UNDERWATER acoustic echo-ranging systems are used extensively[1] in the form of vertical-beam echo-sounders for navigational and surveying purposes and for fish-location, and in the form of horizontal-beam asdics for fish-location and (especially during the Second World War) for the detection of submarines. In these systems considerable success is achieved in the simpler objectives of determining the depth of a level sea-bottom beneath a ship, of determining the depth of a fish shoal provided it happens to be under the ship, and—specially in the case of horizontal-beam systems—even in searching for fish-shoals. But the performance of all these existing systems is severely restricted by the fact that they are single-beam systems. A new system which we and our colleagues have developed retains most of the characteristics of the existing devices, but gives simultaneous information over a wide sector of search with good angular and range resolution. It therefore greatly aids the use of the echo-ranging system for searching, and speeds up and improves the accuracy of survey work. Moreover, it can eliminate the need for mechanical movement of the often heavy electro-acoustic transducer and thus greatly reduce the cost of ship-fitting operations.

In the existing single-beam systems the pulse is transmitted by a transducer (or loudspeaker), and received, after reflexion from the sea-bottom or whale or other object, by the same or a different transducer (this time used as a microphone). The time difference between transmitted and received pulses measures the range of the bottom or object, and is recorded on a chart by a stylus moving at a speed equal to the velocity of sound in water multiplied by the ratio of the width of the chart to twice the maximum range of detection. The transmission of the pulse marks the left-hand edge of the paper, and any echo received causes a mark at a point in the stylus sweep corresponding to the range of the object causing the echo. The paper is meanwhile moving at right angles to the stylus sweep, so that the next pulse transmission marks on a line adjacent to the previous one. The repetition of the echo in the same (or almost the same) position on successive sweeps causes a very great increase in the detectability of the echo if it is a weak one, and if the ship carrying the equipment is moving, the profile of the sea-bottom, or the shape of any object in the beam, is slowly traced out. To look in different directions means rotating the transducer(s), and while such a facility is usually provided in horizontal-beam asdics, it is not usually provided in echo-sounders.

The new system has the following distinctive features: (*a*) The receiving acoustic beam is relatively narrow and is swung continuously over a large sector by electronic means; the rate of sweep is so high that the sector is covered within a time less than the duration of the transmitted pulse. Thus, if the transmitting beam covers the whole sector, then echoes arising anywhere in the sector are received during each pulse repetition interval. (*b*) The received information is displayed on a cathode-ray tube in which one axis of deflexion corresponds to bearing (by being synchronized to the beam-swinging) and the other to range. The intensity of the spot indicates the strength of the echo.

This electronic sector-scanning system clearly enables the shape of the bottom profile over a certain length, or of any object in the sector, to be displayed instantly. The advantages of this in fish-location (and in identification of the fish) and in navigation and surveying may be considerable, and may be judged to some extent from the photographs of actual displays which accompany this article.

Outline of System

A schematic diagram of the system is shown in Fig. 1. The receiving transducer is n times the length of the transmitting transducer, and is divided into n sections, where n is the number of beam-widths (as measured between points where the power response

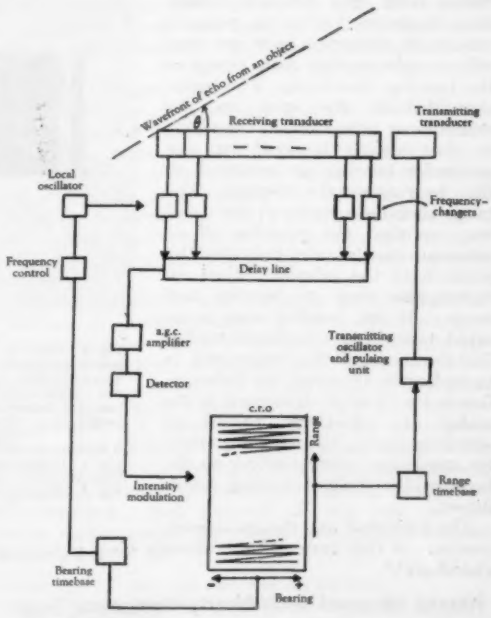

Fig. 1. Schematic diagram of electronic sector scanning system

has fallen to half) it is desired to contain in the scanned sector. If these n sections were connected to n corresponding, uniformly spaced, taps on the delay line, then the beam would be deflected by an amount dependent on the phase-shift in the delay line. The principle of this is readily seen: if an echo is received from a target on a bearing of θ relative to the perpendicular axis of the transducer, then the wave-front of the echo pulse lies parallel to the line shown in Fig. 1. The echo is received by the first transducer section before it reaches the second, and so on. Thus the echo signal received by successive sections is delayed in phase relative to the previous sections. If a peak output is to be obtained for this particular angle of arrival, then the delay line is required to insert compensatory phase-shift so that all the components of the combined output are in the same phase.

In the scanning system, frequency-changer equipment is inserted between the transducer sections and the delay line. The local oscillator which feeds all the frequency-changers is swept in frequency by the bearing time-base, so that the signal frequency received by the delay line varies over a range during every sweep of the bearing time-base. If then the delay line is made to have a phase-shift which varies over this frequency-range from negative values to positive values, the beam will be swept from left to right during each sweep of the bearing time-base. The latter also deflects the spot on the cathode-ray tube from left to right, so that signals received on any particular bearing are recorded on that bearing on the display. The range time-base works in the usual way, so that the position of an echo-spot on the tube indicates the position of the echoing object on rectangular axes of bearing and range. If the bearing scan is so rapid that it is completed within the duration of the pulse and is immediately repeated, no information is lost, and all directions in the sector are effectively looked at simultaneously, but with the angular resolution corresponding to the beam-width of the receiving transducer.

The technical and design considerations of this system have already been published elsewhere[2,3].

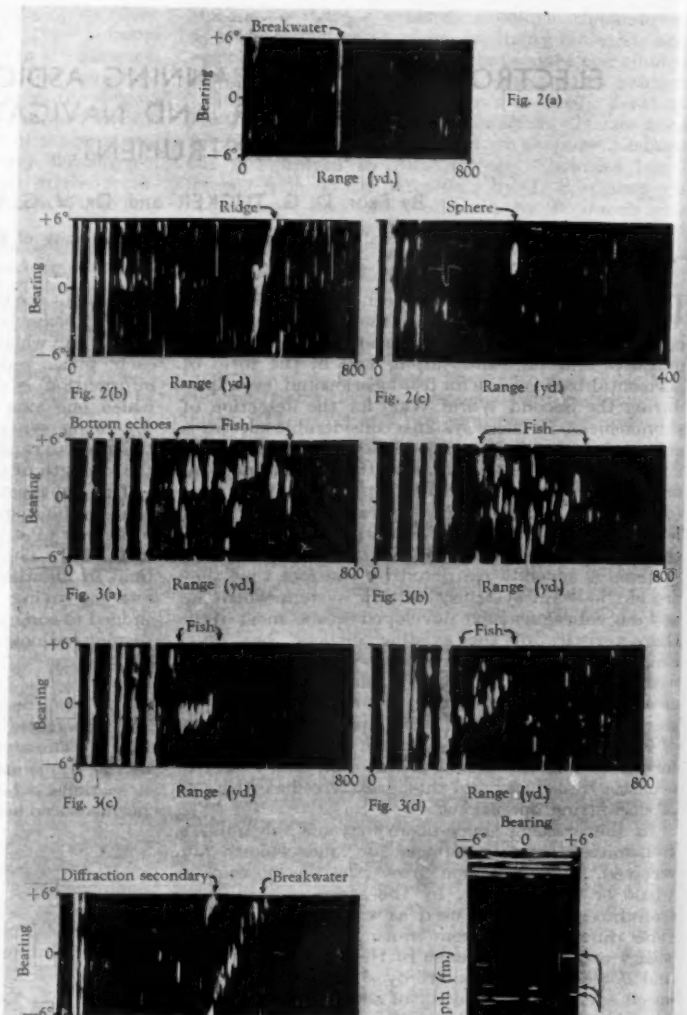

Fig. 2. Some photographs of sector displays taken on R.R.S. *Discovery II*. Beam nearly horizontal (tilt 5° for *a*, *b* and *c*). *a*, Portland Harbour, eastern breakwater viewed exactly at right angles. (N.B. The target echo was so strong that the brilliance was turned down and the echoes from the sea-bottom are lost.) *b*, Ridge on sea-bottom near the Dorset coast. *c*, 18-in. diam. sphere in mid-water at range of 150 yd. in Weymouth Bay. (Several successive traces have been superposed in the photograph as the sphere was moved out by a small boat.) 400-yd. range scale

Fig. 3, *a*, *b*, *c* and *d*. Small fish shoals in southern North Sea. (800 yd. range scale)

Fig. 4. Photograph of full sector display of outer breakwater of Portland Harbour viewed from the east; showing effect of 'side-lobe interference' due to a 'diffraction secondary'

Fig. 5. Scanning echo-sounder display with fish passing through sector. (Southern North Sea, depth about 20 fathoms, max. range on scale = 25 fathoms)

Results obtained with Nearly Horizontal Beam

The first laboratory experimental equipment, made in the University of Birmingham, was, through the courtesy and co-operation of the National Institute of Oceanography, given sea-trials in R.R.S. *Discovery II* during October 1958 and November 1959. On the latter occasion the equipment was used as a research tool by the staff of the Fisheries Laboratory, Lowestoft, in a study of fish migration. The ship was

already equipped with a suitable electro-acoustic transducer designed and constructed by the National Institute of Oceanography. The primary object of the equipment and the trials was the location and study of fish shoals—some small fish shoals are shown on the display in Fig. 3*a*, *b*, *c* and *d*—but photographs of the way in which objects of navigational and surveying interest appear on the display were also obtained. A selection of these latter is shown in Fig. 2*a*, *b* and *c*. Fish can be distinguished from fixed objects, of course, by their movement and the variation of the shape of the shoals. For example, the shoal in Fig. 3*d* is the same as that in Fig 3*c*, but observed a minute or two later. The general speckled background in the photographs is the reverberation, or general back-scattering of sound from the sea-bed and waves. This, being random, never repeats from one pulse to another, and genuine targets can be easily distinguished, not only by their brightness, but also by their consistency from pulse to pulse.

The parameters of the equipment were as follows :

Beam-width = 1·5° in horizontal plane (between ½-power points)
Scanned sector = 12° in horizontal plane, that is, 8 beam-widths
Vertical beam-width = 10°
Pulse duration = 1 m.sec.
Frequency = 37 kc./s.
Transmitted acoustic power = 100 watts during pulse

A technical description of the equipment has been published elsewhere[2].

It should be noted that the heavy lines across the display in the range 0–100 yards approximately are due to the various side-lobes of the vertical beam-pattern striking the sea-bottom at ranges depending on their angle relative to the main beam. In general, no object on the bottom can be reliably detected until after the last of these lines. In an operational system the side-lobes of the vertical beam-pattern would probably be largely removed by suitable transducer design, and the early part of the range, up to the point where the main beam reaches the bottom, would be left clear for the detection of objects in mid-water. But in the experimental trials, the transducer was simultaneously being used for other experimental work which required the side-lobes to be present. The position of the beam in the vertical plane was, however, kept constant by a stabilizing mechanism using a gyro as reference, so that the angle of tilt of the main axis (quoted in Fig. 2) is a known constant for any series of observations.

The fact that there are only 8 beam-widths in the scanned sector in the experimental equipment accounts for the fact that even a 'point' object, such as the 18-in.-diam. sphere, extends over at least one-eighth of the bearing scan. An operational system might with advantage scan more than 20 or more beam-widths.

It is shown in a previous paper[2] how, at the edges of the scan, a 'target' just moving off at one edge begins to show at the other, but as movement proceeds further, the false echo extends with ever-decreasing strength. This is not serious for most purposes, but can be avoided by sacrificing some of the scan and placing a mask over the edges of the display. An illustration of the effect is shown in Fig. 4. It may be possible to get better results by more complex transducer arrangements, and this is being investigated. The effect is in some ways similar to 'side-lobe interference' in radar, but is more correctly known as 'diffraction secondary interference'.

The maximum useful range of detection is of the order 800–1,000 yards at the frequency chosen for the experimental equipment.

Use with Vertical Beam

When the transducer is tilted so that the beam points vertically downwards, the equipment becomes a scanning echo-sounder. If the beam is wide from fore to aft, but narrow from side to side, and scans from side to side, then the display will show, for each pulse transmission, the sea-bottom profile in some detail across the track of the ship. The ship's own motion forwards enables the profile along the track to be recorded. Thus *one* traverse of an area enables a three-dimensional picture of the bottom to be obtained—a result which shows an enormous saving of cost and time in surveying an area. It is, of course, the slant range which is shown on the display, and a correction must be made for true depth.

The vertical-beam operation is also useful for fish location and identification. Fig. 5 shows a photograph of the display obtained when a very scattered shoal of fish (probably herring) was passing through the sector. Individual fish are probably indicated by the smallest of the more distinct marks.

Conclusions

The results obtained by the biologists studying the movements of the fish will be published separately. It can be said, however, that the equipment has already proved itself able to give information on the horizontal and vertical distribution of the fish, and more particularly on their movements, which has probably not been obtained before ; and it may well prove an important research instrument in fisheries and hydrographic work. A system with much higher resolution in range and bearing is being developed, and it may eventually prove possible to study the movements of individual fish, of nets and other objects ; the need already exists.

To summarize, it is thought that the new electronic sector-scanning asdic has considerable possibilities in : (*a*) detection and location of fish ; (*b*) identification of fish by their shoaling habits, echo-strength and shape, etc. ; (*c*) research in fisheries work, for example, in studying the structure and internal and overall movements of fish shoals, and even of individual fish ; (*d*) navigation ; (*e*) hydrographic surveying.

We are grateful to our colleagues in the Electrical Engineering Department at the University of Birmingham for much of the research, design and constructional work which has been involved, and to the National Institute of Oceanography for some financial assistance, for help with the trials, and for the use of R.R.S. *Discovery II* and of the transducer. The interest taken in the work, the use made of the equipment, and the further financial help given by the Fisheries Laboratory, Lowestoft, together with much encouragement and interest on the part of the Marine Laboratories, Aberdeen, have proved a great stimulus.

[1] Tucker, D. G., *J. Brit. Inst. Rad. Eng.*, **16**, 243 (1956).
[2] Tucker, D. G., Welsby, V. G., and Kendell, B., *J. Brit. Inst. Rad. Eng.*, **18**, 465 (1958).
[3] Tucker, D. G., Welsby, V. G., Kay, L., Tucker, M. J., Stubbs, A. R., and Henderson, J. G., *J. Brit. Inst. Rad. Eng.*, **19**, 681 (1959).

THE HARWELL NEUTRON PROJECT

By Dr. M. J. POOLE and Dr. E. R. RAE

Atomic Energy Research Establishment, Harwell, Didcot, Berks

A LABORATORY has been set up at the Atomic Energy Research Establishment, Harwell[1], to exploit the possibility of using a linear electron accelerator as a pulsed neutron-source for time-of-flight studies. These studies increase our knowledge of highly excited states of compound nuclei.

The first machine to be used in this way produced electrons of energy 3 MeV., and was superseded in 1952 by a 15-MeV. machine, which produced photo-disintegration and photo-fission neutrons from a block of uranium which served as target for the electrons. In that accelerator the duration of the electron pulses could be made as short as 0·1 μsec. by pulsing the electron gun, and the beam current during a pulse was of the order of 25 m.amp. at 13·5 MeV. under full-load conditions (400 pulses/sec.). In these circumstances the rate of production of neutrons during the pulse was 10^{14}–10^{15} neutrons/sec.

The new laboratory has been designed around a much more powerful machine than either of its forerunners, since it is capable of providing a beam current during the pulse of the order of 400 m.amp. at 30 MeV. Using this in conjunction with a natural uranium target, a rate of emission of 2×10^{16} neutrons/sec. is possible. However, a further increase in the available neutron output has been obtained by building a special target (called the neutron booster) which is a multiplying sub-critical uranium-235 assembly, and by this means a ten-fold increase in the neutron output has been obtained. Thus the final rate of production of neutrons during the pulse is 3×10^{17} neutrons/sec. (A factor of 1·5 is accounted for by the difference in the production-rate of primary neutrons in natural uranium and uranium-235.)

In the time-of-flight experiments the neutrons are partly moderated in flat water tanks, 2 cm. thick, placed on three sides of the neutron booster. A layer of boron-10 metal placed between these tanks and the uranium-235 core prevents return of slow neutrons to the uranium-235 and so avoids serious distortion of the pulse shape. Some nine windows lead through the shielding and allow neutrons to pass down evacuated 'flight tubes' arranged radially about the neutron source and so reach the experimental apparatus. In this way the time-of-flight, and so the energy of the neutrons associated with some nuclear process, can be measured by recording the time-interval between the accelerator pulse and the time of arrival of the neutron. The recording of the times of arrival of the neutrons is carried out by multi-channel time analysers, one of which is associated with each individual flight path. The ability to carry out a number of time-of-flight experiments simultaneously in this way is one of the main advantages of a pulsed accelerator over a fast chopper as a pulsed neutron-source. The other advantage is the relative ease with which neutron pulses as short as 0·25 μsec. are obtainable.

Experiments in progress include measurements of neutron partial cross-sections for scattering, capture and fission, in addition to total cross-sections. From these measurements, the parameters of nuclear levels extending for several tens of keV. above the neutron binding energy are extracted. These parameters include the level-spacings, the neutron, radiation and fission widths, and the angular momenta. Another type of experiment is the study of the hard γ-rays arising from the capture of a neutron into various levels of the compound nucleus above the binding energy. An analysis of the relative strengths of these energetic γ-rays in different levels in the same compound nucleus demonstrates the random fluctuations that occur in the strength of a particular γ-ray transition.

The new machine should make possible a much more thorough investigation of the statistical laws governing the distributions of the parameters of nuclear levels, and may help to clear up certain mysteries which still surround the phenomenon of nuclear fission. In addition to these studies of neutron interactions, the investigation of a variety of photo-nuclear phenomena is also planned.

In the field of reactor physics, the pulsed-source method has been extended to allow the measurement of neutron spectra in reactor lattices. A knowledge of these spectra is of great importance in calculating the detailed performance of reactors, particularly the phenomena which govern the amount of permissible burn-up. Calculation is in principle possible once a detailed knowledge of the neutron-scattering law is known; but neither experimental results nor methods of calculation are yet sufficiently developed to make this feasible, and recourse has to be made to direct experiment. In this method the target of the pulsed machine is placed inside a section of the lattice of interest, and the neutrons are moderated down to the spectrum characteristic of that lattice. This spectrum is then measured by the usual time-of-flight method.

The increased intensity from the new accelerator will make possible the performance of more complex experiments in which a pulse of neutrons is introduced into a moderator and the spectrum measured by using a synchronized neutron shutter (chopper) which opens a known time after the electron pulse. In this way it will be possible to follow the change in neutron spectrum during the last stages of slowing down, and to make a direct comparison with some of the theories of neutron thermalization available. It will also make possible the direct observation of the so-called 'neutron cooling', that is, the reduction in mean energy of neutrons in a moderator due to preferential escape of fast neutrons.

[1] Poole, M. J., Wiblin, E. R., *et al.*, Proc. Second Int. Conf. on Peaceful Uses of Atomic Energy, Geneva, 1958 (Paper *P*/59).

FORESTRY COMMISSION RESEARCH STATION

BEFORE 1946 the Forestry Commission had no research station; a limited amount of work on silviculture and mensuration was carried out by Forestry Commission staff at headquarters and elsewhere, and work on some other subjects, such as entomology and pathology, was done by specialists attached to other institutions.

In the summer of 1946, Alice Holt Lodge, a moderate-sized country house attached to an old Crown forest, was released from its war-time military occupation and opened as a Forest Research Station. By 1950 this building was proving inadequate for the expanding needs of the Station, and many of the offices were moved into huts. It was obvious that this could only be a temporary measure, and plans for more permanent expansion culminated in the spring of 1959, when a new wing, considerably larger than the building to which it is attached, was ready for occupation. This three-storey wing, together with a single-storey service block and a staff restaurant, has a total floor area of nearly 25,000 sq. ft. Construction is in the form of a concrete framework, supported on piles sunk 36 ft. into the Gault clay. The outer walls are made up of prefabricated panels, containing large timber-framed windows, with timber cladding below. The internal corridors are off-centre, giving rooms 16 ft. deep, suitable for the larger laboratories, on the eastern side, and rooms 12 ft. deep for offices and personal laboratories on the western side. The architects were Messrs. Cowper and Poole, of London.

The two buildings now serve as headquarters for all the research work of the Forestry Commission, with the exception of that on machinery and on utilization development, which is based on London. A branch station in Edinburgh, controlled from Alice Holt, deals with silviculture, genetics and to some extent management problems in Scotland and the north of England. However, much of the work centred on Alice Holt extends into Scotland. Although a silvicultural nursery, a tree-breeding nursery, several seed-orchards for the production of improved strains of tree seed, two poplar nurseries, a botanical collection of poplars and a pathological test nursery are attached to the Station, Alice Holt forest is not itself a research forest, and the experiments on which the staff of the Research Station are engaged are in forests and nurseries scattered all over England, Scotland and Wales.

Much of the field work is supervised by outstationed research foresters, but nevertheless considerable travelling on the part of the Alice Holt and Edinburgh staff is also involved. In research nurseries or at centres where there is a large number of forest experiments, the Research Branch has its own labour forces, but much of the often considerable work involved in laying down forest experiments is carried out with the aid of the ordinary labour force of the forest concerned. It has been found that the progress and success of the work depend very considerably on the maintenance of a proper balance between the graduate staff planning and analysing experiments and the research foresters responsible for their lay-out, supervision and assessment in the field. There is no doubt that the existence of this body of skilled foresters provides a vital link in forest research, which is lacking, or at any rate insufficient, in some other forest research organizations even in countries where more money is available.

The new wing at Alice Holt houses those research sections which have need of laboratory space. Silviculture, with its subsidiary Sections, Ecology and Soils, occupies the whole of the top floor. Silviculture is concerned with all aspects of the cultivation of forest crops from the seed stage onwards. In the past, emphasis in Britain has inevitably been on nursery problems and on the establishment of plantations; but, as Forestry Commission plantings get older, work is becoming more concerned with matters such as the thinning, pruning and manuring of established plantations. Attention is now being given to methods of regenerating felled woodlands. This poses a number of difficult problems that will become more acute when earlier Commission plantings reach maturity. Felling will, of course, provide opportunities for change of species or variety, so that trials of different species, and in particular of different seed origins within those species, are still being actively pursued.

The Ecology Sub-section is concerned with studies of the site relationships of individual tree species, and with the investigation of sites where the establishment of satisfactory tree crops has proved difficult or impossible. The Soils Sub-section has two overlapping functions, analytical services for other sections and in particular for silviculture, and a programme of research into the nutrition of forest trees. Work has been particularly concentrated on foliar analysis and this is proving an essential part of the proper understanding of the many experiments on forest manuring now in progress.

On the second floor, but still under the control of the Silviculture Section, is the seed laboratory. This has a programme of work on improvements in seed-testing technique and on the storage of seed, but its main activity is the testing of all seed lots bought by the Forestry Commission, and for this purpose it is licensed as a 'private seed testing station'. Much of the seed which it tests is housed in the central Forestry Commission seed store, which occupies one end of the ground-floor of the new wing. This is not a research responsibility, but is housed at Alice Holt for convenience, and so that advice on storage and testing facilities are readily available. The store has three large refrigerated chambers, two for routine storage at 2° C. and one at −5° C. for the more difficult species. The total capacity of these chambers is 42,500 lb. of conifer seed. All the conifer seed bought from abroad or collected in England and Wales is stored at Alice Holt.

One end of the second floor is occupied by the Genetics Section. This could be more correctly described as a 'Tree Breeding' Section, since it is concerned mainly with practical problems on the improvement of forest trees. The selection of 'plus' trees from existing stands has already led to the

formation of seed-orchards of Scots pine, Douglas fir, European larch and a few other species. Hybridization, followed by further selection, is being used to effect still further improvements.

The remainder of the second floor is occupied by the Pathology Section, which is concerned with the study and control of tree diseases. The work includes not only damage by fungi and bacteria, but also that due to non-living agencies, such as frost. At the moment, the control of *Fomes annosus*, the most serious cause of butt rot in conifers in Britain, is the major project of this Section. Other projects are top-dying of Norway spruces, a physiological disorder associated with increased exposure of the tree crown, and the testing of poplar and elm clones for resistance to bacterial canker and elm disease, respectively.

The Entomology Section, on the ground-floor, is engaged on investigations of a number of forest pests. It is normally concerned with the biology, including the occurrence of parasites, the forest status, which usually involves population sampling, and the control of each insect. In some cases, for example that of the pine looper moth (*Bupalus piniarius*), work has been particularly directed to finding the population-level at which the insect becomes a potential danger. In another case, that of the larch sawfly, work is directed to finding the effects of different populations and, therefore, of different degrees of defoliation, on tree growth.

The remainder of the ground-floor is occupied by the Photographic Section, which provides a common service to the rest of the Station, as well as to the Forestry Commission as a whole, taking most of the photographs required for illustration and record, and the slides necessary for lecturing purposes. The collection of more than 12,000 black-and-white photographs and nearly 7,000 colour slides is arranged numerically, but fully card-indexed on the International Forest Decimal Classification, so that photographs or slides on any particular subject can easily be located. The Section has produced several short films on forestry subjects.

The old building houses Administration, the Management Section, the Statistics Section, the Library, and a large room, which is used for meetings, conferences, lectures, etc.

The Management Section includes mensuration, which studies the growth of individual trees and of crops, and the methods of measuring them. It produces volume tables, yield tables, etc., for use in forest management. Management also includes the Census of Woodlands, and therefore has charge of all the data resulting from the census of all woodlands of 5 acres or more, carried out between 1947 and 1949, and of the census of hedgerow and park timber carried out in 1951. Working from this basis, sampling surveys for general purposes and local surveys for special purposes will be used in the future to provide more up-to-date information. Now that the hitherto overriding need for a strategic reserve of timber has become less acute, it is obviously desirable that more attention should be given to the economic background of forestry in Britain. For this reason, the Management Section now includes an economist, who is studying the nature and volume of the demand for timber and relating these to the costs involved in raising it. The Management Section is also responsible for assisting the Forestry Commission field officers in the preparation of forest working plans.

The Statistics Section is again a common service to the whole Station. The statistician advises on the layout of all experiments, thus ensuring that the resultant data are suitable for computation. The long-term nature of, and the large areas involved in, many forest experiments make care over design of particular importance. Computation is now done on a 'Pegasus' electronic computer by arrangement with the Royal Aircraft Establishment at Farnborough. The Section also carries out research into the application of statistical methods to problems of forest research and management.

The Library is a small one, containing some 3,500 books; but it receives all the principal forestry periodicals and has a considerable collection of reprints, reports, etc. It also carries out documentation work, resulting in a large card index of literature references on British forestry and on aspects of forestry abroad which are of interest to British foresters. There is also a series of classified information files. All filing is arranged according to the International Forest Decimal Classification.

The Forestry Commission encourages more fundamental work on some of its problems by means of grants to universities and other institutions. This has enabled the Alice Holt Research Station to concentrate the efforts of its staff very largely on the more immediately practical aspects of forest research. Thus, despite the relatively small staff—36 research officers and 107 research foresters in the Research Branch as a whole—there is a considerable output of work, mostly published in Forestry Commission publications (*Forest Records*, *Bulletins*, etc.) or in forestry and other scientific journals. A summary of the work of the Station, with brief accounts of grant-aided work at other institutions and with a number of short papers by the Research Station staff, is published annually as a Forestry Commission Report on Forest Research.

OBITUARIES

Prof. J. G. Wood

The sudden death of Prof. J. G. Wood on December 8 last at the comparatively early age of fifty-nine has not only deprived the University of Adelaide of its senior professor but also has been a severe loss to biological work in Australia.

Joseph Garnett Wood was born at Mitcham, South Australia, on September 2, 1900, and educated at the University of Adelaide, where he obtained his first degree in the honours school of chemistry (1922). Fortunately he had shown interest and ability in botany during his undergraduate course, and in 1923 he became a demonstrator in botany. A programme of work on the vegetation of arid South Australia was beginning and the assistance of a chemist was vital. The saltbush (*Atriplex vesicaria*) which covers huge plains in the arid parts of the State had been termed a halophyte on insufficient evidence. Wood collaborated in the ecological work, which showed that the soils were not saline, except for the scattered salinas where *Atriplex* was absent. The plant had a

high content of sodium chloride in the cells of its uncuticularized, tomentose leaves. Wood investigated the transpiration-rates of a number of trees and shrubs growing in the region and later showed that, in addition to having a low transpiration-rate, *Atriplex* was able to take up moisture through its leaves from saturated air. This work led to the award of an 1851 Exhibition scholarship, and Wood proceeded to Cambridge (Caius College) to read for a Ph.D. On his return to Adelaide in 1927 he was appointed lecturer, soon to be in charge of the Department. He was promoted to professor in 1935.

Work on the vegetation of arid South Australia had led to the generous donation to the University of an area of some 1,000 acres and small field laboratory at Koonamore, about 200 miles north of Adelaide in 1926. Wood collaborated in the work of the Koonamore Vegetation Reserve almost from the start. It is a tribute to his abiding interest in the arid flora that the Reserve and laboratory are still in regular use by the Adelaide Department for teaching and research.

Wood's contributions to botany fall under the heads of general ecology (his work on "The Vegetation of South Australia" (1937) has long been out of print), autecological work on arid plants and nutritional studies. Recently (1957) he and one of his research students have shown that sodium chloride is an essential micronutrient for *Atriplex vesicaria*. The nutritional studies have dealt with nitrogen and sulphur metabolism and the role of such micronutrients as copper, zinc and molybdenum. These are essential for crop production over large areas of leached soils in Australia but are not needed in the same quantities by the native sclerophyllous plants.

Wood had considerable organizing abilities. The fine range of biological buildings at the University owes much to his planning. Possessed of an alert mind and known for his fairness and even temper, much of his time and energy of late years have been spent on committee work. He was a member of the Interim Council of the Australian National University during 1948–51 and a Council member during 1952–55. He served on the Advisory Committee of the Commonwealth Scientific and Industrial Research Organization during 1951–59 and was chairman of the Board of Standards, of the Australian Journals of Scientific Research, and chairman of the Board of Research Studies of his University at the time of his death.

His recreations were largely connected with his home and garden. He had a good knowledge of Australian contemporary art and he himself painted. In 1930 he married Joan Hazel, who, with their three daughters, survives him. His one-time teacher and colleague can only join his many other friends in deploring his sudden passing.

T. G. B. Osborn

Prof. W. R. Fearon

William Robert Fearon, professor of biochemistry in the University of Dublin, died on December 27, 1959, aged sixty-seven years. The only child of a Presbyterian clergyman who had died when his son was four years old, he entered Trinity College, Dublin, in 1911. It was an indication of the intellectual versatility which was to characterize his life that he found some difficulty in deciding whether to read English or science. In the event he read natural science, in which he graduated B.A. with a Senior Moderatorship in 1915, securing also a Foundation Scholarship in Experimental Science two years later. He entered the Medical School during this period, but became so intensely interested in physiology and biochemistry that he interrupted his medical studies in the fourth year to do research work in nutrition in London during the First World War. Following this, he was awarded an exhibition to Emmanuel College, Cambridge, where he became one of the devoted band which Frederick Gowland Hopkins gathered around himself in the University Biochemical Laboratory. On his return to Trinity College, Dublin, in 1921, he was elected to a fellowship and thus began a period of almost forty years during which he enhanced the reputation of the College with his researches, delighted his students with the brilliance of his lecturing and entertained his colleagues by his sparkling conversation on almost any topic.

His literary bent had found recognition while he was still a student in the award of the vice-chancellor's prize in English verse, the University Philosophical Society's Gold Medal for English composition and the editorship of the College miscellany, *T.C.D.*, to which he was a frequent contributor. Throughout his career his clear, limpid style was to stamp everything he wrote whether scientific or otherwise. He was a master of the happy descriptive phrase which left an indelible impression. Always interested in motor-cars, he wrote of a visit to the Lancia showrooms in London during the early '30's that he had been taken for a drive in a car "that was the size of a cottage and cost as much as a house".

His early work was concerned with the formation of urea from protein, in the course of which he began to exhibit that facility for developing colour reactions which was to lead to so many papers in the *Biochemical Journal*. He loved to demonstrate these reactions to his colleagues or to visitors, and he must have discovered far more than he ever published. His interest in nutrition persisted and found its most recent expression in a survey of the fluoride content of Irish drinking waters. It was typical of his alert mind that he began this work more than ten years ago, at a time when others were only commencing vaguely to realize the possible practical significance of fluoride intake in dental health. For about six years before his death he had been interested in the ureides obtained from autolysed yeast. An outstanding contribution was his book, "An Introduction to Biochemistry", first published in 1934; he was working on the fourth edition when he last became ill. It is remarkable in that it is at once "An Introduction" which unfolds the subject to the student systematically and with the utmost clarity, and a comprehensive review that can be consulted with profit by the more mature worker.

Notwithstanding a rather shy disposition, he had wide interests outside his laboratory and, indeed, outside College. In the '20's he was the friend and associate of the literary and artistic giants of the time in Dublin—W. B. Yeats, Jack Yeats, George Russell (AE), Lennox Robinson, Oliver St. John Gogarty. He held the honorary professorship of chemistry of the Royal Hibernian Academy. His play, "Parnell of Avondale", was a success when

produced at the Abbey Theatre nearly twenty years ago. He was an accomplished pianist and organist, and had an extensive knowledge of music.

One of the fascinations of knowing Fearon was the frequency with which, even after years of close friendship, one discovered new interests of his: a large stamp collection, to which he added regularly; his flair for cars, both vintage and modern sports—his own collection had included a Lancia, a Bentley and a Rolls; his real appreciation of the musical comedies of the early part of this century. In the same way, one learned with surprise, in 1943, that he was a candidate for a seat in the Senate of the Irish Parliament as a representative of the University of Dublin, for one had never been aware of his interest in matters political. Yet he continued to hold his seat to the end, and his colleagues in the Upper Chamber often expressed a high regard for him as a member.

William Fearon was a man of great gentleness of character but one who did not encourage intimacy, and not many of his friends knew him really well. Nevertheless, his passing will leave many gaps. He will be missed in the College Common Room and in his clubs as in the class-room and in the Senate chamber.

W. J. E. Jessop

NEWS and VIEWS

Experimental Physics at Liverpool : Prof. J. M. Cassels, F.R.S.

Prof. J. M. Cassels, who has retired from the chair of physics in the University of Liverpool, joined the staff as a lecturer in 1952. In 1955 he was promoted to senior lecturer and in 1956 was appointed to the professorship of experimental physics which had been instituted a few years before but had remained vacant. He had gone to the Atomic Energy Research Establishment as a research fellow in 1949 on completing his Ph.D. at Cambridge having previously done some war work there. At Harwell, he joined the research group working on the new 170-MeV. synchrocyclotron and played a leading part in the experimental programme on p–p and n–p interactions. He transferred to Liverpool to work with the higher energy (380 MeV.) Liverpool cyclotron, which came into full operation at the end of 1954. Largely because of Cassels's foresight, they had a full experimental programme ready to go, and this has continued at a steady pace ever since. Cassels ran a small group of his own, and his name is on many of the laboratory's papers, especially some concerned with pion interactions. But his influence went far beyond his own group, and inspired much of the high-energy work of the laboratory. His strength is both in experimental techniques and in his grasp of theory. He is also an enthusiastic teacher, and a very fine lecturer. He resigned his post to take up, in the autumn of 1959, a visiting professorship in Cornell University, where he will be able to continue his work on high-energy nuclear physics. In 1959 he was elected a Fellow of the Royal Society.

Dr. A. W. Merrison

Dr. Merrison, who has been appointed as Prof. Cassels's successor, was educated at King's College, London, and after taking a degree went to work on centimetric radar at the Signals Research Development Establishment. In 1946 he transferred to the Atomic Energy Research Establishment, Harwell, and worked on various aspects of neutron spectroscopy in the Nuclear Physics Division. He was appointed as lecturer in the University of Liverpool in 1951 and transferred to the research appointment known as the Leverhulme lectureship in 1954. He became a group leader on work with the large 380 MeV. synchrocyclotron and published several important papers on pion physics. In the spring of 1957 he was appointed to a staff post at CERN, the international high-energy laboratory at Geneva. There he worked with the 680-MeV. synchrocyclotron which started work shortly after. One of the results of his team, the discovery of the rare mode of decay of a pion directly into an electron instead of through the normal muon, is considered as of exceptional importance in confirming the theorists' general conception of these particles. Recently, Merrison has been concerned with arranging work for the 28-GeV. proton synchrotron at CERN which has just started working. Dr. Merrison had no opportunity of taking a normal Ph.D. course in a university on account of the War, and so took his degree as a member of the University of Liverpool staff. He is expected to return to the University in the autumn of this year.

Geological Survey and Museum : Sir William Pugh, O.B.E., F.R.S.

Sir William Pugh, director of the Geological Survey of Great Britain and the Museum of Practical Geology, is retiring at the end of July. Sir William, who was born on July 28, 1892, was educated at the County School, Welshpool, and at the University College of Wales, Aberystwyth. After service in the First World War, in which he was made O.B.E. (Military Division), received the French Croix de Guerre and was twice mentioned in dispatches, he was appointed in 1919 professor of geology at Aberystwyth. In 1931 he was appointed to the chair of geology in the University of Manchester, and became pro-vice-chancellor and later deputy vice-chancellor of that University. He was awarded the Murchison Medal of the Geological Society in 1952 for his researches into the stratigraphy of the Palæozoic rocks in Central Wales. Sir William became director of the Geological Survey and Museum in 1950. He was elected a Fellow of the Royal Society in 1951, and knighted in 1956.

Dr. Cyril J. Stubblefield, F.R.S.

Dr. Cyril J. Stubblefield is to succeed Sir William Pugh as director of the Geological Survey of Great Britain and the Museum of Practical Geology. Dr. Stubblefield was educated at the Perse School, Chelsea Polytechnic and the Royal College of Science, London. In 1923 he was appointed demonstrator in geology in the Imperial College of Science and Technology, a post he occupied until 1928, when he joined the Geological Survey as a geologist. In 1929 he entered the Department of Palæontology of the Survey; and in 1947 he was appointed chief palæontologist. This post he held until 1953, when

he was promoted to an assistant directorship in charge of Special Services, and Northern Ireland. Dr. Stubblefield has made many important contributions to our knowledge of the palæontology and stratigraphy of the Palæozoic rocks, and in particular has made a special study of the trilobites. His contributions to geological science have received recognition from the Geological Society of London, by the award of the Bigsby Medal in 1941, and, in 1955, the Murchison Medal. He was elected to the Royal Society in 1944. Dr. Stubblefield's geological activities have not been restricted to his work on the Geological Survey, for he has been an active member of various scientific societies. He is at present president of the Geological Society of London, vice-president of the Palæontographical Society, and a member of the Council of the British Association.

Willis Rodney Whiting Award : Dr. Morris Cohen

DR. MORRIS COHEN, of the National Research Council, Ottawa, is to be awarded the Willis Rodney Whiting Award for outstanding work on corrosion. The presentation will be made in Dallas, Texas, on March 14 by the U.S. National Association of Corrosion Engineers. Dr. Cohen is head of the Corrosion Group in the Division of Applied Chemistry, Canadian National Research Council. His work has been largely in physical chemistry, finding what forms corrosion takes, identifying corrosion products, determining rates of corrosion in pure metals and metal alloys. The investigations have included the polishing, etching and electroplating of metal surfaces in order to study the effects of such treatment on the resistance of the metals. Dr. Cohen was born at Regina in 1915, received his B.A. degree at Brandon College in 1934, and his M.A. and Ph.D. degrees at the University of Toronto. He joined the staff of the National Research Council in 1943.

U.S. Navy's Bathyscaphe *Trieste*

ON January 24 the U.S. Navy's bathyscaphe *Trieste* successfully dived to a depth of 37,800 ft. in the Pacific Ocean. The region selected for the dive was the Marianas Trench, the depth of which had previously been estimated at 5,364 fathoms (32,184 ft.). Pressure measurements on the bathyscaphe reached 16,883 lb. per sq. in. during the half-hour stay on the ocean floor. The total descent was accomplished in 4 hr. 42 min., and the ascent in 3 hr. 17 min. The operation was directed by M. Jacques Piccard and Lieut. Don Walsh.

New Research Qualification

THE Royal Institute of Chemistry has established a Research Diploma for work in pure or applied chemistry to be awarded to graduate members, associates or Fellows of the Institute under conditions similar to those for the Ph.D. degree of a university. The Research Diploma will be particularly directed to those who are unable to satisfy the requirements for taking a higher degree of a university, but who may be able to undertake research under an approved supervisor in a technical college, an industrial firm, a Government department or a research association. The scheme will apply equally to university graduates and those who qualified at technical colleges. Further information can be obtained from the Assistant Registrar, Royal Institute of Chemistry, 30 Russell Square, London, W.C.1.

International Computer Federation

ELEVEN nations have now ratified the status of the newly-formed International Computer Federation. The Federation was formed following the first international conference on Information Processing, sponsored by Unesco and held in Paris during June 1959. At a provisional committee for the Federation the following members were named : *Chairman*, Isaac L. Auerbach (United States) ; *Vice-chairman*, Prof. A. A. Dorodnicyn (U.S.S.R.) and A. van Wijngaarden (Netherlands) ; and *Secretary*, J. A. Mussard (Unesco). It is expected that the first meeting of the Council of the Federation will take place later this year, resulting in plans for a second international conference on information processing, with an associated technical exhibit in 1963. Representation of the United Kingdom is by the British Computer Society, Ltd., Finsbury Court, Finsbury Pavement, London, E.C.2, from which further information can be obtained.

First All-India Congress of Zoology

AT the first All-India Congress of Zoology, sponsored by the Zoological Society of India and held at the University of Jabalpur during October 24–28, the president, Dr. M. L. Roonwal, director of the Zoological Survey of India, referred in his address to the great number of species comprising the fauna of India and pointed out how little is known of the biology of many of even the commoner ones. He also directed attention to the rapidly decreasing numbers of some of the larger animals and appealed for more effort in conservation. Turning to field ecology, he emphasized what a wide field is available for research which can be done with the aid of a microscope and quite simple equipment. He added, however, that what the worker has to create for himself is a suitable mental atmosphere which will enable him to do hard, arduous work, perhaps for years on end, and without thought of rushing into print too soon merely to increase the number of his published papers. Dr. Roonwal concluded with a plea for the training of more systematists to work on the national zoological collections in the charge of the Zoological Survey of India. The Congress which followed was divided into six sections at which more than a hundred papers were presented, and symposia on marine zoology and fishes in the Indian Ocean and on the re-orientation of teaching of zoology in India were held.

The Coryndon Memorial Museum, Nairobi

DESPITE the fact that Dr. L. S. B. Leakey, curator of the Coryndon Memorial Museum, has been heavily engaged with work on the Olduvai human remains and lectures in Britain and elsewhere, steady progress is recorded in the annual report for 1958. Probably the two most important exhibits during the year were the Shakespeare Second Folio and living examples of rare snakes. The former is placed in a special case and pages are turned periodically. The live snakes form a major attraction and included gaboon viper, rhinoceros horned viper, boomelang, garter snake and the rare Storms' water cobra. Another exhibit was that of the striped weasel which is only the second to be recorded in Kenya. With the giant human teeth from Olduvai and a Chellean I stage of culture, an ivory tool from the Chellean II level and some samples of red-ochre used by hand-axe man are shown. It is interesting to note that a new method of skinning small mammals has been

devised, and a paper on the process is in course of preparation.

Palæoliths at Leopoldville

H. van Moorsel, keeper of the prehistoric and geological collections at the Lovanium University at Leopoldville, has published an article in the Faculty of Sciences on the palæolithic finds in the neighbourhood of the city. There is a vast stony plain in the vicinity which seemingly is very ancient, as it does not appear to have any connexion with the existing river terrace systems. Archæological finds have been few in number, but the author claims to have recognized Sangoan and Middle Stone Age tools. It is difficult to judge from reproductions of photographs and possibly some of the suggested tools figured are really naturally fractured, but there is no reason to think that these cultures were not represented. The article was read as a paper at the fifth Congress of Prehistoric and Protohistoric Sciences at Hamburg in 1958.

Central Electricity Generating Board Research Fellowship

The Central Electricity Generating Board is offering a research fellowship, tenable at St. Catherine's College, University of Oxford, in the physical sciences, including metallurgy, engineering or mathematics. The fellowship, tenable for three years, will take effect from October and will be valued at £1,000 per annum. It is open to persons of British nationality, who by the date of his or her appointment have taken the degree of doctor of philosophy or gained the equivalent experience in research. Applications should reach the Master elect, St. Catherine's College Planning Office, 42 Banbury Road, Oxford, not later than March 31.

Refresher Course on Science in Schools

The Department of Extra-Mural Studies and Education of the University of Durham, King's College, Newcastle, is organizing, in conjunction with the North-Eastern Association of the British Federation of University Women, a refresher course on Science in Schools. The course is intended primarily for women with scientific training who, having had to interrupt their careers, now wish to take up or return to teaching in schools. The course will take place at the University during March 23–30. Further information can be obtained from Miss C. Cruse, 292 Ovington Grove, Newcastle upon Tyne, 5.

University News : Belfast

The following appointments are announced: Dr. D. J. Carr (Melbourne), to the chair of botany; Dr. R. A. Henstock, to be senior lecturer in pure mathematics; and Dr. A. L. Stewart, to be lecturer in applied mathematics.

Monash

The following first appointments of staff at Monash University, the newly established University in Victoria, Australia, are announced: professor of biology, Dr. A. J. Marshall, reader in zoology and comparative anatomy at St. Bartholomew's Hospital Medical College, London; and professor of chemistry, Dr. R. D. Brown, senior lecturer in general chemistry, University of Melbourne.

Announcements

Sir Patrick Linstead, rector of the Imperial College of Science and Technology, University of London, has been appointed chairman of the Science Advisory Committee of the British Council and a member of the Executive Committee of the Council in succession to the late Sir Alfred Egerton.

Sir Harold Roxbee Cox, vice-chairman of the National Council for Technological Awards, has been appointed chairman in succession to Lord Hives, who has retired. Sir Harold Roxbee Cox, formerly chief scientist, Ministry of Fuel and Power, is deputy chairman and chairman-elect of the Metal Box Co., Ltd., and a director of Wilmot Breedon (Holdings), Ltd., and of Boulton Paul Aircraft, Ltd.; he is a member of the Council for Scientific and Industrial Research.

The Scottish Branch of Aslib is organizing its annual general business meeting and a one-day conference to be held on February 5 in the Scottish Central Library, Lawnmarket, Edinburgh. Further information can be obtained from Miss N. I. Shinie, T. and H. Smith, Ltd., Edinburgh 11.

The twelfth international symposium on Crop Protection is to be held at the Agricultural College of the University of Ghent on April 26. The symposium will coincide with the twenty-fourth five-yearly international Floralia of Ghent. Further details can be obtained from Prof. J. van den Brande, State Agricultural College, Ghent, Belgium.

The joint annual meetings of the Chemical Society and the Royal Institute of Chemistry will be held in Belfast during April 5–8 at the David Keir Building of Queen's University, Belfast. Further details can be obtained from the Joint Annual Meetings Officer, Royal Institute of Chemistry, 30 Russell Square, London, W.C.1.

The spring meeting of the Clay Minerals Group of the Mineralogical Society will be held during April 12–13 in the Department of Chemistry at the University of Aberdeen. The topic for discussion will be "Clay Minerals in Sediments". Further details can be obtained from Dr. R. M. S. Perrin, School of Agriculture, University of Cambridge.

Three courses of six lectures each will be given at the Borough Polytechnic, Borough Road, London, S.E.1, on the following topics: Variety Reduction in Engineering (commencing on February 17); Lubrication and Lubricants (commencing on February 22); Foundry Sands (commencing on February 9). Further information can be obtained from the Secretary of the Polytechnic.

The Association of Oil and Colour Chemists has announced that the twelfth Technical Exhibition will be held at the Royal Horticultural Society's New Hall, Greycoat and Elverton Streets, London, S.W.1, during March 15–17. Further details can be obtained from the secretary, Mr. R. H. Hamblin, Association of Oil and Colour Chemists, Wax Chandler's Hall, Gresham Hall, London, E.C.2.

Prof. A. Masevich, vice-president of the Astronomical Council, U.S.S.R. Academy of Sciences, Moscow, is to give a number of lectures throughout Great Britain on "Conquest of Space". These lectures are being organized by the Society for Cultural Relations with the U.S.S.R. at the Royal Festival Hall, London, February 6; Birmingham Town Hall, February 8; Colston Hall, Bristol, February 9; and the Free Trade Hall, Manchester, February 12. Tickets are available from the agents of the various halls.

SHELL DEPARTMENT OF CHEMICAL ENGINEERING, CAMBRIDGE

IN March 1945 the University of Cambridge gratefully accepted the offer of the Shell Group of Oil Companies to found a School of Chemical Engineering. This generous gift amounted to £435,400 and was accompanied by an offer to make an annual sum of £2,500 (later increased to £3,000) available until further notice, thereby forming the Shell Chemical Engineering Studies Fund.

The first Shell professor of chemical engineering, T. R. C. Fox, took up his appointment in October 1946 and teaching started in October 1948. Since that time 187 students have graduated in chemical engineering and the teaching staff has grown to its present establishment of six lecturers, three demonstrators and an assistant director of research (two of these posts are vacant at present).

The teaching in the Department consists of a two-year course taken either after Part I of the Natural Sciences Tripos or Part I of the Mechanical Sciences Tripos. This course leads to the Chemical Engineering Tripos, normally taken in the fourth year. From the outset it has been the aim of the Department to provide a scientifically based training both for those who have been led to the subject through their interest in chemistry and for those whose primary training has been in engineering. It is not the intention to mould these two groups into a single pattern but rather to modify both and let them temper one another. In this way the Department has pioneered a new development in chemical engineering education.

Graduates in chemistry or engineering from other universities may also be admitted to the course, which qualifies them for the B.A. degree.

In the early days the Department was housed in a temporary single-storey building in Tennis Court Road; eventually 9,000 sq. ft. of temporary accommodation was provided on this site. After the exploration of many alternative schemes, a permanent site adjacent to the Cavendish, Metallurgy and Zoology Laboratories was allocated in 1955. Mr. J. Murray Easton was appointed architect and a building with a net floor area of 22,000 sq. ft. was designed to replace and incorporate parts of the old chemical laboratory. Work on the building started in November 1956 and the Department began to move into the first section of its new building early in 1959. The main group of laboratories was opened by Lord Godber, chairman of the Shell Group of Companies, on December 9, 1959 (Fig. 1).

The Laboratories of the Department are partly in a new steel-framed building of five floors and partly in the four-storey building along the Pembroke Street frontage, originally built in 1907, which has been virtually rebuilt internally. The last stage of the rehabilitation of the 1907 building (the ground floor now occupied by the Chemistry Laboratory) will begin in June 1960. The two buildings have been welded into a whole by a new staircase which feeds floors in the converted building from half landings.

The basic design envisaged relatively large-scale experiments being carried out at the lowest level (in what the Department terms the Unit Operations Laboratory (Fig. 2), which is 48 ft. × 45 ft. × 22 ft. high) with adjacent service workshops, stores and instrument laboratory. On the first floor are centred the teaching activities with lecture rooms, library, chemical laboratory and cloakroom. On the second and third floors are eleven combined office-research laboratories for teaching staff, five small research laboratories and one large research laboratory for chemical bench work. Existing structural and service limitations restricted the use of the converted 1907 building. A bank of six constant-temperature rooms (four having 16–17 ft. headroom) are built in the region of the junction between the two buildings, and on the first floor advantage was taken of the headroom to provide a ramped lecture theatre capable of seating both first- and second-year classes together. The basement is used for stores and mechanical services and a new roof structure was built to provide a drawing office and common-room in place of a group of attics at various levels.

In the Unit Operations Laboratory which passes through two floors there are galleries to permit access to apparatus at 4 ft. and 11 ft. above floor-level. There is also a central well 5 ft. deep and provision in five places to pass through the floor of the laboratory above, so increasing the effective ceiling height at these points from 22 ft. to 35 ft.

Fig. 1. New building for the Department of Chemical Engineering, Cambridge

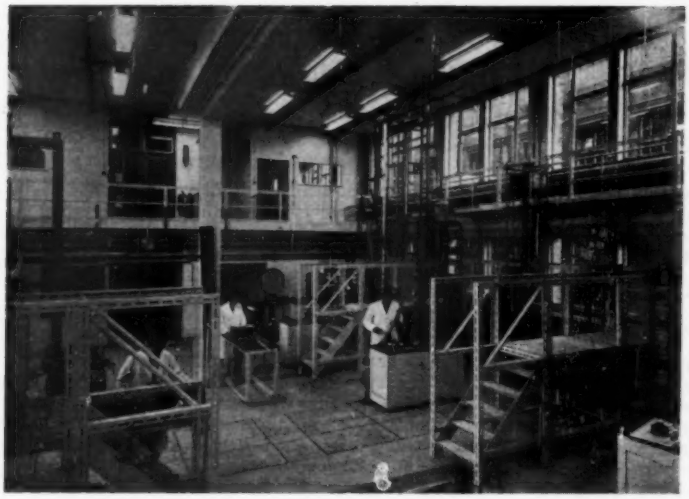

Fig. 2. Unit Operations Laboratory

Each experimental position in the laboratory has steam (10 and 100 lb./sq. in.), compressed air (10 and 100 lb./sq. in.), gas, water and electrical (60 amp.) services. These services are provided by: (1) two steam ring mains operating at variable pressures and supplied by three boilers with a combined capacity of 2,500 lb./hr.; (2) two compressed air ring mains (one 5 in. in diameter for part of its length) operating at variable pressures up to 100 lb./sq. in. and supplied by four reciprocating compressors providing up to 540 c.f.m.; (3) two water ring mains fed from a 6,000 gal. capacity tank in the roof. In the case of both air and steam there is a cross-over pipe network to permit any compressor or boiler to feed either of the pair of ring mains. Additionally there is a general ventilation extract system, balanced by a warmed air intake, and there is also a furnace extract system.

Fume cupboards in the research laboratories have been installed in the centre of the building in spaces between ducts. The fume cupboards are of the 'walk in' type and as the tray at bench height is removable they may be used for high towers and similar apparatus. They are finished internally with an epoxy resin. The cupboards are arranged in vertical banks of three, each bank being exhausted by a single fan. Two of these banks have interconnecting, lined trunking to permit the installation of apparatus approximately 6 in. × 12 in. through three floors, that is to say, approximately 40 ft. high in what becomes effectively a single fume cupboard. Provision has also been made to install apparatus if required in ducts (5 ft. 6 in. × 3 ft. 6 in.) which run almost the full height of the building. These ducts are now fitted with removable floors and used as cupboards.

In the earlier years of the Department's existence, research work necessarily took second place to the development of the teaching programme. However, its importance has steadily increased, and to date twenty-one research students have submitted work for research degrees.

A feature of the Tripos course is that students carry out a modest research project during their final year, rather than a formal laboratory course in chemical engineering operations. Some information of genuine value has been obtained.

The industries or other fields of employment entered by graduates on leaving are as follows:

Petroleum and petrochemicals	29
Chemicals (excluding petrochemicals)	80
Chemical plant manufacture	21
Atomic energy	23
Miscellaneous	34

The original staff of the department is now widely dispersed. Prof. K. G. Denbigh was the first professor of chemical technology at Edinburgh, Prof. E. S. Sellers became the first professor of chemical engineering at Swansea, Prof. J. M. Kay is at the Imperial College, London, and Prof. P. V. Danckwerts has recently returned from the Imperial College to take up the Shell professorship following the resignation of Prof. T. R. C. Fox, who is to continue teaching work in Cambridge in the Engineering Laboratory.

THE SCIENCE MASTERS' ASSOCIATION

NEARLY seven hundred science teachers from many parts of the British Isles gathered for the annual meeting of the Science Masters' Association, which was held in the University of Southampton during December 29, 1959–January 1, 1960.

There was a large attendance at the Guildhall on the evening of December 29, when members of the Association were welcomed to Southampton by the Mayor, Mrs. R. M. Stonehouse. The civic reception was followed by a lecture, illustrated in colour, on "Southampton Borough Archives" by the Borough archivist, Mr. Bruce Jones, and members were able to examine some of the historic treasures of Southampton, which were displayed with models and illustrations of modern educational buildings prepared by the Borough architect, Mr. L. Berger.

The programme included lectures and demonstrations, visits, discussions, films and, in addition to the usual members', manufacturers' and publishers' exhibitions, a number of special exhibitions which attracted considerable interest.

The morning sessions were devoted to lectures and discussions which fully maintained the high standards of the annual meeting. Of special interest to physicists were the lectures by Prof. A. M. Taylor on "Some Demonstrations in Light and Colour" (which

filled the large physics lecture theatre to overflowing on two successive mornings), Mr. S. Weintroub on "The Solid State", Mr. B. H. Venning on "Analogue Computers and their Use as a Teaching Aid", and Mr. E. Wolfendale on "The Junction Transistor and its Application". Chemists enjoyed lectures by Dr. G. W. A. Fowles on "Directed Valency" and Dr. H. M. Frey on "Reactions of 'Hot' Molecules", while the interests of biologists were stimulated by lectures by Prof. W. T. Williams on "The Applications of the Pegasus Digital Computer to the Solution of Problems in Plant Ecology and Taxonomy", by Dr. K. A. Munday on "Modern Developments in the Use of Cooling in Physiological and Biochemical Studies", and by Mr. T. H. Blank on "Research on Game Birds". Many teachers found pleasure and profit in lectures not immediately connected with their own branches of science, notably in hearing Prof. E. J. Richards on "Some New Problems in Aeronautical Research", Dr. N. S. Grassam on "Nuclear Engineering—Past, Present and Future", and Dr. K. Hecht on "Atomic Physics in West German Schools".

A most interesting programme was arranged for afternoon visits to industrial establishments, various departments of the University, and places of general interest. The traditional exhibitions were well supported; the sixty manufacturing firms represented had put on a fine display and reported exceptionally keen interest in their products, while there was a steady stream of members to see the excellent publishers' stands in the pleasant setting of the Lanchester Engineering Building. The members' exhibition, always a popular focal point at the annual meeting, and housed this year in three laboratories in the Department of Physics, showed all the usual fertile ingenuity and craftsmanship that is an object lesson and a stimulus to those who are not so skilful or so inventive.

An important feature of the meeting was the special exhibition on "Atomic and Molecular Structure and Nuclear Energy", with some fine displays on this theme by firms and other organizations in the United Kingdom, the United States and West Germany. These included a student reactor which employs natural uranium fuel with a light-water moderator and a small polonium–beryllium neutron source, a reactor simulator and monitoring equipment, and many pieces of apparatus and teaching aids which would be valuable to schools, technical institutions and universities. Equipment from America included a micro-wave analogue of the Bragg diffraction apparatus, using aluminium balls to simulate the crystal lattice structure, an ingenious α-particle counter, and a neat apparatus for the determination of Planck's constant. There was an impressive display of equipment which is available for the teaching of atomic physics in West German schools. Other special exhibitions included visual aids, a feature display on textile research, and apparatus and teaching aids contributed by teachers in secondary modern schools.

During the meeting simultaneous discussions took place on draft syllabuses in physics, chemistry and biology, prepared by subject panels set up by the Association to implement the policy statement, "Science and Education", which was published in 1957. A fourth panel is working on a syllabus for education in science as part of general studies in the sixth form.

A most enjoyable discussion by the Science and Religion Group of the Association was initiated by Dr. Donald MacKay, of King's College, London, who took as his theme "The Christian Doctrine of Man".

A real highlight of the Southampton meeting was touched on Wednesday evening, when Dr. D. G. James, vice-chancellor of the University, gave his presidential address to a packed audience in the refectory of the University, following the Association dinner and University reception. Dr. James, speaking on the subject of "Literature and Science", emphasized the imperative need to bring the sciences and the humanities together in education; calling for more intellectual excitement and the necessity to look beyond one's own special interests to try to see them in relation to others and to the abiding questions, he concluded: "It cannot be doubted that, as boys and girls in ever-increasing thousands move on to the sixth form and the universities, the enjoyment of literature and the arts by the majority who will read science may not be sacrificed except at a severe cost. At the same time, it is clear that the life of science shares with the arts the power of the creative imagination, and that fellowship of the scientist and the artist, instead of being, if Sir Charles Snow is right, a rare thing, should be the most natural thing in the world".

The marked growth of the Association in membership and in its links with other organizations was a major topic at the business meeting on December 31.

From a membership of 2,700 in 1945, the number had grown to 4,500 in 1955 and is now 6,500, the growth continuing at a steadily increasing rate. This rapid development has thrown an increasing burden on the officers of the Association, and the chairman, Dr. H. F. Boulind, emphasized the urgent need for a permanent headquarters with salaried staff to enable the work of the Association to be carried on effectively.

Regarding the links of the Association with other bodies, it was pointed out that, with the growing realization of the significance of science in education and the consequent activity by many organizations and individuals, there was bound to be some duplication of effort through ignorance of what others were doing, some waste of resources and some inefficiency. This indicated an urgent need for a co-ordination centre as a home for science education. This would at least reduce some of the overlapping and wasteful use of personal and financial resources; more positively, this venture could have a really valuable effect on science education not only in Britain but also more widely, for example, through Unesco and the Organization for European Economic Co-operation, through associations of science teachers in other countries and through individuals using the Centre. The Association already has many valuable links with other organizations both in Britain and abroad; others are being forged, and the meeting was very glad to welcome Prof. H. F. Meiners, of New York, who had come to Southampton to discuss the extension of the happy co-operation between the Association and the American Association of Physics Teachers. The Science Masters' Association is sponsoring the establishment of the proposed Science Education Centre in January 1960 as one way of celebrating the diamond jubilee of its foundation. The Centre is being set up temporarily at Holt, in Norfolk, but it is hoped that sufficient financial and other resources will be put at the disposal of the Association to provide more adequate accommodation and equipment in a less-remote spot in the near future.

H. LOCKETT

ECOLOGY OF PARASITES

THE autumn meeting of the Parasitology Group (Institute of Biology) was held in London at Birkbeck College on October 30, 1959. In the opening discussion of the meeting, which was entitled "The Ecology of Parasites", Prof. B. G. Peters (Imperial College, London) considered the effect of soil factors on plant parasitic nematodes. Soil-living stages are directly influenced by edaphic factors such as moisture, temperature, chemical constitution of the soil and the presence of predators and of possible hosts. Prof. Peters went on to point out that in the parasitic phases, complex relationships exist between the biomasses of host plants and parasitic eelworms. With increasing plant nutrients in the soil there is an increase in host biomass and a correlated increase in parasite biomass. However, at a fixed nutrient-level, the correlation between host and parasite biomasses is negative and the parasites retard the development of their hosts. In species of *Heterodera* having one generation each season, the multiplication rate is highest at low population-levels. As the parasite population density builds up so the rate of multiplication falls. Concurrently, the host at a fixed population density shows a correlated fall in biomass until it can barely maintain the eelworm population. Under these conditions, catastrophic crop losses occur in one season and can alternate with reasonable crop yields the next.

Dr. R. Lainson (London School of Hygiene and Tropical Medicine) spoke about the adaptations shown by coccidial parasites to their environment. The life-histories of sporozoan Protozoa show striking modifications which may be regarded as allowing the parasite to overcome the hazards of transmission and which form an evolutionary pattern, often serving as a basis of classification.

Eimeria, a typical coccidian, has one host in its life-cycle and the difficulties of transfer are overcome by the production of resistant oocysts, which can remain viable outside the host for long periods. The hæmogregarines have eliminated the dangers of a free-living stage and are passively transferred by ectoparasitic invertebrates such as leeches, ticks and mites. The parasites circulating in the blood cells of the vertebrate are ingested by a blood-feeding invertebrate, which itself is eaten by a new vertebrate host.

Yet other coccidians are further adapted and undergo most of their life-cycle in the ectoparasite, the vertebrate virtually serving as a reservoir of infection for the invertebrate host. Besides these modifications in the mechanism of transmission, changes have taken place in the processes of fertilization and sporogony, and all these adaptations ensure maximum efficiency in the maintenance and dissemination of the species.

Dr. S. D. Garrett (Botany School, Cambridge) discussed inoculum potential. Infectivity denotes the potentiality for infection of a single infective organism. In order to produce disease in 50 per cent of inoculated host individuals a large dose of infective agents is often required, and the term 'inoculum potential' denotes the infection potential of this dose. The necessity for the large dose in establishing infection can be explained either by the low probability for infection by a single organism, the hypothesis of independent action, or by the necessity for synergic action between individual infective organisms—the hypothesis of synergistic action.

Evidence available from plant pathology supports that from animal pathogens and accords better with the hypothesis of independent action. However, the possibility still exists that synergism may be involved in the production of disease in an 'unnatural' host, for example, where it is necessary to apply heavy doses of rust fungus spores to the leaf surface to produce disease. The chief evidence of synergistic action is provided by experiments with fungi infecting the roots of trees. The high resistance of the roots can only be overcome by a fungal inoculum provided with a food base, and infection is accomplished by mycelial strands and not by individual hyphæ.

Dr. G. Salt (King's College, Cambridge) spoke about the environment of larval parasitoid insects. The ecology of these parasitoids has two distinct aspects. The adults are free-living, usually winged, and the ways in which the females select the hosts and parasitize them have been widely studied. However, the ecology of the parasitoid egg or larva living on or in a particular host has received relatively little attention. Dr. Salt described some of his work on the life-history of a parasitoid larva within the hæmocœl of its larval insect host and gave an account of the environment, the changes which must occur in it if the parasite is to survive and the interactions between host and larva. An 'unnatural' host reacts to the presence of the larva in its hæmocœl, the larva becoming surrounded by an envelope of host blood which prevents development.

Dr. D. Arthur (King's College, London) described the ways in which ticks select their hosts and the feeding sites upon them. Hard ticks, on the tips of vegetation, are aroused to a state of preparedness by non-directional, tactile stimuli. These stimuli are reinforced by interacting odour and temperature stimuli prior to attachment to the host. In soft ticks, temperature appears to be the dominant stimulus for attachment.

The range of hosts selected by the parasite is not precisely known because of incorrect identification of ticks by earlier workers. However, the majority of ticks appear to be non-specific in their host selection. Some are limited to certain groups of hosts, but very few are specific to one host species, even among primitive ticks feeding on primitive mammals and the more advanced forms such as those found exclusively on Primates. For example, *Ornithodorus moubata* appears in three biological forms, that found on warthogs, porcupines and antbears possibly being primitive. The sites of attachment of various species and the reasons for their selection were discussed.

The chairman, Dr. E. L. Taylor (Veterinary Laboratory, Weybridge), opened the discussion. The adaptations of a parasite show various aspects

operating simultaneously. The ecology of stages outside the host is similar to that for free-living organisms, but the parasite has also to find its host, effect an entrance or attachment and then behave in a way that ensures its own and its host's continued existence. The effect of parasite on host has been widely studied, but the effect of the host on the parasite should also be considered. Indeed, the two physiologies are interacting and in some circumstances mutual benefit may accrue. Contrary to the usual tenet of successful parasitism, the parasitoid insect larva kills its host, and Dr. Salt commented that the percentage of host population infected was small and losses, which could be considered as due to a refined form of predation, could easily be supported. Dr. Hindle cited examples of the extinction of host species by parasitism. Immunity reactions of the host, although prejudicial to an individual parasite, operate for the benefit of the species by limiting the parasite population. This mechanism is different in plant and animal hosts, and Dr. Jones said that plants do not show immune reactions, but rather a resistance to infestation which is a property of the individual cells. Parasite populations of plants show both density-dependent effects and independent factors, such as the steady rate of decay of the population of cyst-forming eelworms in the soil.

Immunity in invertebrates has been little studied, but there appears to be no true immunity although the production of non-specific antibodies may be evoked by the introduction of any foreign object. When the parasitoid insect larva with its envelope of host blood is transferred to another similar host there is no homograft reaction such as occurs in vertebrates.

Dr. Garrett suggested that another important difference between the plant and animal host is that in the plant there is always a region of young tissue at the growing points, whereas in animals growth usually ceases at maturity. Young animals are more susceptible than adults, and in plants the rootlets, for example, are more susceptible than the roots, the resistance of which can only be overcome by the most specialized of parasites. This phenomenon of age-resistance also operates in favour of the parasite by limiting the population.

The proceedings of this meeting will not be published in full.

JUNE MAHON

ORIGIN OF TEKTITES

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CERTAIN aspects of recently advanced concepts relating to the origin of tektites require some considerable modification in the light of the conclusions arising from long-continued and detailed studies of the tektites from Australia. So far as is known, australites are the most recently arrived tektites on the Earth, and have evidently not lain upon the Earth's surface for more than 5,000 years. Furthermore, in terms of preservation of original shape types modified by secondary phenomena generated during rapid transit through the Earth's atmosphere, they are unmatched by any tektites from any other strewn-field on the Earth. Consequently, they are in the best condition on which to base conclusions as to their origin. Moreover, the very best preserved, most perfectly developed australites, which come from the Port Campbell district on the southern coast of Western Victoria, are retained in Australia, and have not been critically examined by overseas writers who make reference to australites in their publications.

Much detailed information has now accrued relating to the primary shapes, the secondarily modified shapes arising from the action of aerodynamical phenomena during ultra-supersonic and supersonic flight through the Earth's atmosphere, and the effects of tertiary phenomena such as subaerial erosion resulting from 5,000 years exposure to atmospheric agents. Considerable and detailed studies of the specific gravity of the same and different types of shape from the same and from different localities have been carried out. Many refractive index determinations have been made and specific refractivities calculated. Arcs and radii of curvature of remnant primary (posterior) and newly formed secondary (anterior) surfaces have been determined graphically from silhouette tracings, and significant conclusions drawn from these. Several etch solution tests have been conducted which provide valuable information of the effects of natural etching upon original structural elements and sculpture patterns of australites. Three more chemical analyses have been made and the composition related to specific gravity and refractive index values. The effects of heating to 1,200° C. has added further knowledge to the australite problem generally.

Studies of Darwin glass, 'straw-silica glass' and other non-volcanic terrestrial glasses have led to the rejection of Darwin glass from the group of the true tektites. Thin-section studies of the internal schlieren of australites have clarified the relationships between the core or body portion and the circumferential flange. All the structures, sculpture patterns and present shapes of australites can be adequately explained in terms of the few primary forms developed in an extraterrestrial environment, and subsequent modification by aerodynamical phenomena during high-speed earthward flight through the Earth's atmosphere, with a further modification by fragmentation, abrasion and solution-etching of the shapes so developed.

The results of these and other detailed studies have appeared during the past two decades[1].

The question of the origin of australites is admittedly still unsolved, but the weight of evidence from Australian studies is strongly in favour of an extra-terrestrial source.

The concept has recently been advanced[2] that the arrival of tektites from space, with the distribution found on Earth, is not possible. Urey has calculated

that if australites arrived as a swarm, its mean density must have been greater than 10^{-6} gm. cm.$^{-3}$, its diameter some 10^{8} cm., and hence there would be a piling up of australites to a depth of 100 gm. cm.$^{-2}$ over southern Australia. Urey further states that he knows no answer to this problem. Assuming that the mean density and diameter of the swarm of australites are those given by Urey, detailed studies of australites and their distribution immediately supply the answer to the problem of their piling up to the depth suggested. As Urey states, this piling up is certainly "not the case". In fact, such a piling up to the depth calculated never could occur, for three very significant processes have operated to prevent it. The first and most important process operated for a very short period of time, but was, indeed, most effective in reducing the bulk of the swarm. The australites provide all the necessary evidence to show that they entered the Earth's atmosphere as small, cold (and hence solid) bodies with pre-formed primary shapes and pre-determined sculpture patterns, and that during high-speed transit through the atmosphere, in a matter of minutes at most, many of these forms were reduced in bulk by up to and more than 65 per cent, some by up to 80 per cent. This arose from the development of shock waves causing high pressures and temperatures, the generation of laminar boundary layer flow and reversed boundary layer flow leading to fusion stripping and ablation of the leading surfaces of the australites, and consequently considerable reduction in the size of each original form. Here, then, is an excellent means of reducing the estimated figure of a piling up process by two-thirds and more, and the amount lost in this way must be even greater when it is considered that an untold number of smaller original forms (less than 0·5 cm. in diameter) were undoubtedly completely consumed during atmospheric flight, as evidenced by studies of the smallest complete forms that survived the processes of ablation and thin-film fusion-stripping.

Urey has expressed surprise that a small glass object 1–2 cm. in diameter would travel at high velocity through the atmosphere, keeping one orientation as is necessary in the case of the more button-shaped tektites. Nevertheless, australites have done just this, and, moreover, not the "more button-shaped" examples alone, but also the ovals, boats, dumbbells, teardrops and canoes, etc., the better preserved examples of which all reveal identical types of secondarily developed structures and features (for example, circumferential flanges, concentric flow-ridges, clockwise spiral flow-ridges, anticlockwise spiral flow-ridges, radial flow-lines on the anterior surfaces, and so on). The role of aerodynamical phenomena at high speeds of flight supplies the answer to the development of all these features, and nearly all the australites bear testimony to the maintenance of a steady state of airflow, a state that can only be achieved during supersonic (and ultrasupersonic) speeds of atmospheric flight, with translatory forward motion only, and neither rotation about a polar axis, nor overturning during flight. As long as supersonic airflow obtains, a state of steady flight is maintained; but evidence is present on the anterior surfaces of some australites to indicate that although the motion of the objects was definitely limited to one direction, there arose some degree of 'wobbling' as an outcome of form-drag and buffeting at the high speeds that prevailed. Whether any of the australites in the swarm were affected by accidental or overtake collisions, it has not yet been possible to prove.

After australites landed on the Earth's surface, many without fracturing on impact, processes of abrasion and solution-etching played their part throughout the next 5,000 years or so, thus further reducing the mass of many forms. This is confirmed from detailed studies of the curvature – size relationships of excellently preserved australites, taken in conjunction with studies of other australites that reveal a whole range of tertiary modifications by subaerial agencies, from slightly worn and etched shapes that are almost perfect, right through to strongly abraded and/or deeply etched, now incomplete specimens, and all manner of fragments that can be assigned to one or another of the few shape groups, as well as many nondescript fragments. On the whole, australites have not lost anywhere near the amounts of glass by abrasion and etching as have tektites from other strewn-fields in other parts of the world, such as bediasites (which are very much older in terms of the time they have been on earth surface) and rizalites, etc. (which are also older, but also occur in regions subject to more pluvial, tropical conditions). Nevertheless, significant amounts of abrasion and solution-etching have been effective in certain parts of the Australian strewn-field, more especially in the arid regions and marginal lands, as is evident from comparisons between specimens from those regions and specimens in better preserved states from the more temperate regions of the island continent.

However, it is at present impracticable to assess the amount of australite glass lost from abraded specimens or the amount removed in solution by etching, for the whole of the 2 million square mile strewn-field. All that can be said with certainty at present is that the examination of some 20,000 specimens of australites in various states of preservation from various localities within the strewn-field indicates that between 50 per cent and 75 per cent of these have lost some 5–10 per cent of the original form, some by attrition from sand-blasting in certain areas, others from rolling in stream beds, some by solution-etching in certain soils, some by both processes, according to their location in the vast strewn-field. Several specimens of australites that are badly worn or deeply etched have lost 50 per cent or more of the original form. Furthermore, studies of the etching of smaller forms and small fragments lead to the conclusion that an unknown number has been completely destroyed by processes of subaerial erosion.

Current etching experiments with australites reveal that a specimen weighing approximately .2 gm. passes completely into solution in 4 per cent hydrogen fluoride in approximately 1,180 hr. at room temperatures. The rate of solubility was 3–4 mgm./hr. during the initial 300 hr. immersion, falling to less than 0·5 mgm./hr. after 1,000 hr. immersion, at which stage the initial form had been reduced in bulk by approximately 99 per cent. Although soil solutions are expected to be far less potent than 4 per cent hydrogen fluoride, nevertheless such solutions have been operative in varying concentrations and in varying quantities from place to place for some 5,000 years. In the most favourable circumstances, therefore, it is apparent that some australites in the right environment where solution-etching has been prevalent have been much reduced in bulk as a

consequence of the removal of soluble ions. That entire forms of australites can have been completely dissolved by such a process is indicated by the 2-gm. weight australite that dissolved in 4 per cent hydrogen fluoride in 1,180 hr. without leaving any insoluble residue whatsoever.

Here, then, is a second important factor responsible for causing australite distribution to appear as it does to-day, and it is thus a significant factor in further reducing Urey's estimate of what the thickness of australites should be on the Earth's surface if entering from outer space as a shower; originally the thickness was somewhat greater than now, but loss by subaerial agents of erosion acting for some 5,000 years has been of considerable importance.

A third factor of possibly lesser importance than the first two, but nevertheless of some significance, affects the present distribution of australites on the surface, and hence, indirectly, the thickness of the australite 'pile up' from place to place. This factor introduces the operation of transporting agents such as streams, birds and aboriginal man. It has been generally accepted in the past that australites are normally found in the places where they originally fell to Earth. This seems to apply in many parts of the strewn-field, but in others there is indisputable evidence of movement by stream action into alluvial tin and alluvial gold deposits, as well as into clay pans and lake beds. I have observed several that have been washed over the edges of steep marine cliffs by surface run-off; these fall into talus cones and beach deposits and are ultimately lost by abrasion or swept out by tides on to the adjoining sea-floor. Others have been observed in stream beds entering the sea at grade, and thus provide a pointer to the fact that many specimens that become washed into water-courses ultimately are carried out on to the sea floor from farther inland.

It is a well-known fact that the Australian emu and the plain turkey utilize australites of suitable shape and dimensions as gizzard stones, and thereby can transport australites over considerable distances, at the same time subjecting them to marked degrees of wear in their function as 'grinder-stones'; these birds have been doing this sort of thing since australites first became available to them on the surface of the Earth, as evidenced by the discovery of gizzard stones with the fossilized remains of their ancestors. Aboriginal man has further added to the problem of present australite distribution, inasmuch as he prized these tektites as objects of magic and mystery, and carried them around on his nomadic journeyings; this is further evidenced by the discovery of australites in ancient aboriginal camps and kitchen middens, and by the fact that some show signs of 'carry polish' while others have been fashioned into crude, small implements.

Such events in the post-atmospheric flight history of australites must undoubtedly be taken into account when considering the initial scatter and thickness of australites in the strewn-field. There is no shadow of doubt that in many places the original distribution was by no means what it is to-day.

Thus, the three factors: (*a*) ablation and fusion stripping of primary forms during ultrasupersonic and supersonic earthward flight through the atmosphere; (*b*) subsequent abrasion, fragmentation and solution etching by subaerial agents; (*c*) the habits and activities of certain native birds and aboriginal man; have all been potent in affecting[1] the amount of australite glass that ultimately reached the Earth's surface, and then[2] in affecting its distribution and thickness on the surface. They all add up to a logical reason for the fact that australites are not now piled up to the calculated thickness of 100 gm. cm.$^{-2}$ if they came in from outer space as a shower, and with this objection to the present known thickness removed, added to the knowledge that australites show all the marks of having been subjected to the action of aerodynamical phenomena at high speeds of earthward flight downwards only through the atmosphere, there should be no really serious objection left to the entry of australites from outer space as a shower. Inasmuch as the other tektites from the other known seven strewn-fields of the Earth are much older, much more abraded or infinitely more affected by solution-etching, and do not reveal the remarkable features possessed by australites, it is at present impossible to extend *in toto* these ideas to explain their origin.

Further to the problem of tektite origin, Urey has set out reasons for regarding the several puzzling features of tektites as being capable of explanation in terms of derivation from cometary collision. In fact, however, the reasons advanced only serve to confuse the issue even more than it has been confused in the past several decades. The same arguments apply against the chances of cometary collision being the cause as arguments already advanced[1] against the much earlier theories such as Spencer's meteorite splash hypothesis[3] and Kahn's contraterrene meteorite impact theory[4].

Fatal to all three theories are the facts that the eight accepted types of true tektites fell to the Earth at widely separated times in late- to post-Mesozoic Earth history, and hence cannot possibly fit into the scheme of one cometary collision every 50 million years. The 50 million year cometary cycle was evidently designed to produce bediasites in the Eocene and australites, etc., in the Pleistocene.

The geological evidence, however, shows that the moldavites are Middle Miocene, the Ivory Coast tektites have been suggested to be late Mesozoic, while the bediasites were found in Pleistocene gravels and said to be derived from the Eocene. There are time-intervals of 10–30 million years between these assigned ages. Later in Earth history, the indomalaysianites are regarded as Middle Pliocene, the javaites as Middle Pleistocene, the rizalites as middle or early part of the Late (? Upper) Pleistocene, while the australites are undoubtedly the youngest of all, and definitely Holocene.

There are time-intervals of 3 or 4 million years between the groups of younger tektites, and time-intervals of 15–20 million years between them and the Miocene moldavites. It is thus apparent that the idea of tektite generation in 50 million year cycles of cometary collision cannot be accepted as it stands. Moreover, such cometary collisions would have to result in the meagre total of 2,000 known bediasites by the Eocene cometary collision, produced from a very restricted part of the Earth's surface, and at the other end of a 50-million year cycle, another cometary collision would be required to produce about 600,000 known australites, rizalites, javaites, billitonites and indomalaysianites spread over thousands of square miles of territories widely separated from one another.

Even abrasion, fragmentation and solution-etching differences between the results of the first cometary collision and the second could scarcely be expected

to account for such a discrepancy, and at all events, the big Indomalaysian–Philippines–East Indies–Australian strewn-fields contain tektite types that fell to the Earth at different periods of late Tertiary to Quaternary Earth history. A single cometary collision in the Pleistocene cannot therefore explain the origin of the tektites ranging in age from Middle Pliocene to Holocene in the Far East–Australian region. Such a high-energy explosion as that postulated to have affected so vast an area of the Earth's surface must surely have had greater and more far-reaching effects than the mere production of a few thousand pieces of glass of relatively small size ; no such effects have been recognized or postulated.

A further difficulty with such a suggested origin is that the single high-energy explosion would be required to fuse all manner of terrestrial materials of different composition, so as to produce tektite glass of generally similar chemical composition in areas 65° of latitude apart. Even if the cometary collision supplied the energy for fusion, the wide array of superficial terrestrial materials would by no means supply relatively homogeneous tektite glass, even with the postulated fractionation into acid and basic fractions, a process which, in itself, is most difficult to conceive during the upward propulsion of fused Earth material that afterwards re-entered Earth's atmosphere according to the cometary collision theory. There should, moreover, be a whole range of partially fused materials formed at each milieu affected by the cometary collision ; there should also be a whole range of different kinds of glass irrespective of the postulated acid and basic glasses ; there should be removal of the basic glasses (or else they have never been found), leaving only the acid tektite glass. Furthermore, this one postulated cometary collision would have to be responsible for shaping the different types of tektites in different ways to produce different shape types at widely separated strewn-fields, and would, in some unexplained manner, have to return them to Earth at intervals separated by some 3 or 4 million years. Another cometary collision would then be necessary to explain the Middle Miocene moldavites, and so on, with the result that as many cometary collisions would need to be postulated, and all within the limits of the originally suggested 50 million year cycle of cometary collision, as there has been suggested Great Circles to account for tektite distribution. In other words—a meaningless maze.

Finally, the australites are not consonant with the suggestion of fusion of Earth substance by a high-energy explosion, propulsion to great heights (beyond the atmospheric envelope) with the production of primary forms, and then return to Earth through the atmosphere to generate the secondary shapes.

Admittedly high-speed transit through the Earth's atmosphere has produced the secondary shapes from primary forms. It is not agreed, however, that the composition of australites can be explained in terms of the fusion of the kind of materials exposed at the Earth's surface, and at the same time show the type of chemical gradient according to a provincial distribution across 2,000 miles of the island continent. There is no evidence whatsoever that australites received their primary shapes by upward propulsion from the Earth's surface. None of the known shape types reveal the characteristics shown by volcanic bombs, for example, which are propelled in a plastic state, sometimes to considerable heights, although never, as far as known, high enough to leave the Earth's atmospheric envelope. All the evidence so far accrued is convincing that the australites received their secondary shapes by the modification of primary spheres, spheroids, dumbbells of revolution and apioids of revolution that entered the Earth's atmosphere as solid, non-rotating bodies at ultra-supersonic velocities, traversed the atmosphere at lesser speeds nearer the Earth's surface, passing through a supersonic zone, through the transonic zone and finally into the subsonic zone. During this transit, their leading surfaces were sculptured into the shapes and designs now known from studies of the very excellently preserved, perfectly developed specimens from the Port Campbell district of Western Victoria.

All the evidence points to the fact that the majority of these forms maintained stable positions during high-speed flight through the atmosphere, and while supersonic or higher speeds prevailed, they always presented one surface only to the effects of the aerodynamical phenomena responsible for their secondary sculpture. Moreover, each entity in the swarm, whether original sphere, spheroid, dumbbell or apioid, or even hollow form, behaved in unison with the rest, and each received identical treatment from the medium through which it had its trajectory, so that identical secondary features were produced on the forward surface of each shape type, such features only varying in minor degrees according to the shape types on which they were produced. No secondary shapes have been found that would indicate modification of such geometrically possible forms of revolution as the annular torus and the paraboloid ; but this does not mean that they were not present. Some of the so-called 'aberrant' types of australite shapes, which have been subjected to the secondary modification by aerodynamical phenomena in the same way as the better known shape types, were undoubtedly pre-formed shapes before entry into the Earth's atmosphere from outer space. They may have been originally of the paraboloid or annular torus shape types that suffered accidental collapse or other modification in their extra-terrestrial birthplace.

In the face of all these problems and difficulties that beset research work on tektites, I cannot but conclude that cometary collisions spaced at 50-million year intervals are entirely incapable of satisfactorily explaining tektite origin, or of solving the present distribution of widely spaced strewn-fields containing different shape types of tektites that fell to Earth at widely separated intervals. The position still stands as it did approximately seventy years ago, when it was stated about tektites (then referred to as "obsidianites")—"where they came from no one knows". Research since then has shown that for the australites at least, an extra-terrestrial origin of tektite glass is by far the most likely, but it is still unknown whence in extra-terrestrial space the tektites were derived.

[1] Baker, G., *Proc. Roy. Soc. Vic.*, **49** (2), 165 (1937) ; *Min. Mag.*, **25**, 487 (1940) ; *Proc. Roy. Soc. Vic.*, **52** (2), 312 (1940) ; *Mem. Nat. Mus. Vict.*, **14** (1), 7 (1944) ; **14** (2), 47 (1946) ; *Proc. Roy. Soc. Vic.*, **60**, 35 (1950) ; *Trans. Roy. Soc. Sth. Aust.*, **76**, 1, 27 (1953) ; *Proc. Roy. Soc. Vic.*, **67**, 165 (1955) ; *Min. Mag.*, **30**, 596 (1955) ; *Mem. Nat. Mus. Vict.*, **20**, 59, 173 (1956) ; **21**, 72 (1957) ; **22**, 1 (1957) ; *Amer. J. Sci.*, **256**, 369 (1958) ; *Mem. Nat. Mus. Vict.*, **23**, 5 (1959) ; **24**, 69 (1959). Baker, G., and Forster, H. C., *Amer. J. Sci.*, **241**, 377 (1943). Baker, G., and Gaskin, A. J., *J. Geol.*, **54**, 88 (1946).

[2] Urey, H. C., *Proc. U.S. Nat. Acad. Sci.*, **41**, 27 (1955) ; *Nature*, **179**, 556 (1957).

[3] Spencer, L. J., *Nature*, **131**, 117 (1933).

[4] Kahn, M. A. R., *Contrib. Met. Soc.*, **4** (1), 35 (1947).

[See also p. 300 of this issue.—Editors.]

LIPOAMINO-ACIDS: THE QUESTION OF ARTEFACT

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THE name 'lipoamino-acid' is proposed for a lipid molecule as normally defined[1] from which one or more amino-acid molecules can be cleaved. For lipoamino-acids in which peptide bonding is known to occur the name 'lipopeptide'[2-7] is appropriate.

There are ever more numerous reports of the existence of lipoamino-acids in lipid samples prepared from many biological materials. Samples demonstrably uncontaminated with free amino-acids or peptides afford various amino-acids on acid hydrolysis. The amino-acids appear to be in covalent linkage, since lipoamino-acids have survived at a wide range of *p*H values (without heat) and during treatment with salts under various conditions, solvent fractionation (including countercurrent distribution), electrodialysis, electrophoresis, paper chromatography, and column chromatography on alumina, cellulose, 'Hyflo Supercel', and silicic acid. They have even survived treatment with proteolytic enzymes, Van Slyke deamination[8], and dialysis with *N* hydrochloric acid for three days[9].

One lipoamino-acid, phosphatidylserine, has been isolated pure, and its chemical structure is established. Others have only been revealed as minor constituents of mixtures, which in a few cases contained no phosphorus. Their structures cannot be analogous to that of phosphatidylserine, particularly because in many samples hydroxy amino-acids were absent or were present only in small proportions.

Some samples containing lipoamino-acids were devoid of free amino-groups, as indicated by negative ninhydrin tests. For example, in my hands lipid fractions from blood, egg, flies, liver, and milk, eluted from silicic acid by 5 per cent (v/v) methanol in chloroform, under no conditions gave more than a faint yellow ninhydrin colour on heavily loaded paper chromatograms. The sodium 1,2-naphthoquinone-4-sulphonate test[10] was also negative, yet hydrolysis liberated amino-acids. Total amino-acids (chiefly glutamic) liberated from the fly (*Drosophila melanogaster*) fraction[11] amounted to 0·07 mole per kilogram of lipid. They were liberated at 105–110° by alkali as well as by acid, but not by water, and not at atmospheric temperature and pressure by hydrogen with Adams catalyst.

Peptide bonding could account for absence of free amino-groups, but was ruled out for the above fractions, and for some others, by negative chlorination tests[11] and by absence of 'Amide II' absorption in the infra-red (unpublished experiments).

Detection of amino-acids after hydrolysis of lipid preparations does not *necessarily* indicate the occurrence of peptide or protein moieties, and claims in the literature that it does should be viewed critically.

During examination of lipids from several sources by silicic acid chromatography it was noticed that commercial dried blood and egg, and decaying *Drosophila*, gave fractions with higher and more varied amino-acid content than sources believed free from non-enzymic and/or enzymic deterioration. It therefore seemed possible that lipoamino-acids might be artefacts not existing *in vivo*. Binding of nitrogenous substances is a well-known if ill-understood concomitant of lipid autoxidation[12,13], which might occur during extraction and manipulation.

Experiments with linseed oil supported this hypothesis. Raw linseed oil was freed of phospholipids and other contaminants by passage in chloroform through a silicic acid column. (It still contained minute traces of lipoamino-acids.) This oil (10 ml.) was mixed with water (10 ml.), glycine (0·25 gm.), and hæmoglobin (0·1 gm.) in a large test-tube into which a brisk stream of filtered air was passed for 2 days at 25°. The chloroform-soluble portion of the product was chromatographed on silicic acid. Material eluted by pure chloroform afforded only traces of amino-acids on hydrolysis, but material (ninhydrin-negative) eluted by 10 per cent methanol in chloroform afforded approximately 2 mgm. of glycine. Purification by dialysis, instead of by chromatography, confirmed that silicic acid was not responsible for this result. Unreacted, contaminating glycine was only eluted from the column by pure methanol[11]. Leucine and serine, used in place of glycine, gave similar results. When hæmoglobin was omitted from the reaction mixtures, or when the oil was subjected to normal conditions of lipid extraction, no binding of amino-acids could be detected.

However, being less prone to autoxidation than highly unsaturated phospholipids, linseed oil is perhaps not a good model substance.

The following experiment was therefore made. A solution of glutamic acid uniformly labelled with carbon-14 (156 000 counts/min., weight negligible) in moist *n*-propanol was mixed with powder, prepared from *Drosophila* (15 gm.) and dry ice[11], before propanol extraction. The crude extract contained 105,000 counts, 41,000 being retained in ligroin-soluble material (0·82 gm.) which was then chromatographed on silicic acid (12·5 gm.). Specific activities of eluted fractions (in which > 90 per cent of counts were recovered) were determined using thin samples on aluminium planchettes. Results are shown in Table 1. That radioactivity was bound in lipid was demonstrated by paper electrophoresis[3]. Material eluted by 15 per cent methanol in chloroform was placed on a heavily loaded zone of non-radioactive glutamic and 2-pyrrolidone-5-carboxylic acids before electrophoresis. Afterwards, at least 85 per cent of the counts were found in the lipid zone at the

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origin. Confirmation was provided by 'three-run' paper chromatography (Fig. 1). When lipid eluted by 15 per cent methanol was thus tested zone *U* contained 22 per cent of the counts, *V* + *W* contained 62 per cent, and *X* + *Y* contained 16 per cent. Percentages for lipids eluted by 25 per cent methanol and by 35 per cent methanol were respectively 21, 67, 12 and 27, 30, 43. Zones *U* to *Y* contained fluorescent zones of autoxidation products, which probably accounted for the considerable losses of radioactivity from lipid zones. Significantly, lipid zones did retain more than 20 per cent of their original counts.

Results also shown in Table 1 are of an experiment in which a considerable weight (1·0 gm. for 8 gm. *Drosophila*) of arginine hydrochloride (372,000 counts/min.) was used instead of the negligible weight of glutamic acid.

Clearly, some lipoamino-acids are artefacts formed *in vitro* from endogenous amino-acids; indeed, apart from phosphatidylserine, all of them might be artefacts. This possibility was tested by a series of experiments in which large amounts of selected amino-acids and other compounds ('additives') were added, singly or in pairs, to render concentrations of endogenous compounds insignificant from the beginning of extraction. Extraction and removal of non-lipid contaminants were carried out at low temperature[11].

In a typical experiment blood (50 ml.), collected from a horse, was within 2 min. poured into methanol (100 ml.), plus powdered dry ice (excess), plus additive (10–40 gm. per gm. total lipid). In a room at − 15° lipids were then extracted (total solvent amounted to 400 ml. chloroform–methanol, 2 : 1, v/v) and washed four times by the Folch (1957) sodium chloride

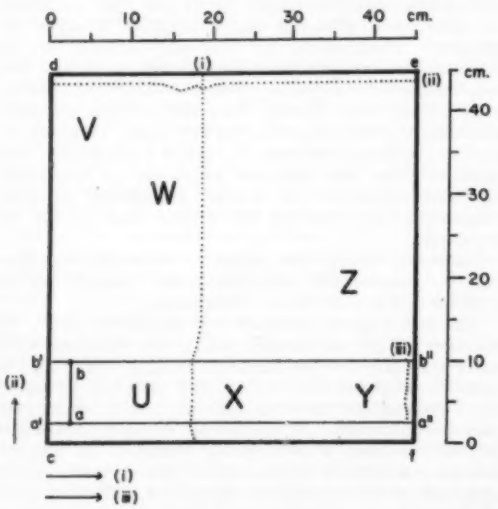

Fig. 1. 'Three-run' paper chromatography, a test for radioactive glutamic acid bound to lipid. Lipid was deposited on a line, *ab*, on a sheet of Whatman No. 3*MM* paper, *cdef*, and chromatographed (i) in benzene/*n*-butanol/methanol/water (1 : 1 : 2 : 1, v/v). After development in the second dimension (ii) with water, the strip *a'a''b'b''* was cut out, and heavy loads of non-radioactive glutamic and 2-pyrrolidone-5-carboxylic acids were applied to *ab* before development (iii) with water. Development was ascending. Numbered, dotted lines represent respective solvent fronts. Lipid formed a zone at *U* and the two acids at *Y*. The paper was treated with ninhydrin, and ultimately cut into several pieces, designated *U*–*Z*. These were eluted with 25 per cent water in *n*-propanol for radioactivity measurements

Table 1. Incorporation of Radioactive Amino-Acids into *Drosophila* Lipids during Extraction and Manipulation

Eluant	Counts/mgm./min. of eluted material	
	Glutamic acid	Arginine
$CHCl_3$	0	0
5 per cent MeOH	9 46	1
15 „ MeOH	7	1 7
25 „ MeOH	7	7
35 „ MeOH	116	7
50 „ MeOH	83	9
MeOH	1,510*	42*

* High values due to elution of the free amino-acids[11].

procedure[11]. After evaporation of solvent (< 40°) the whole lipid (0·17–0·20 gm., all ligoin-soluble) was chromatographed on a silicic acid column (7·5 gm.). Materials eluted successively by chloroform (60 ml.), by 5 per cent (60 ml.), 15 per cent (240 ml.), and 25 per cent methanol in chloroform (150 ml.), and by methanol (80 ml.) were hydrolysed, and the hydrolysates were examined by paper chromatography[11]. Cow's milk, within 3 min. of milking, was handled similarly, except that additive in aqueous solution was mixed with it immediately before pouring into methanol. Whole *Drosophila* were powdered with dry ice plus additive, as mentioned above. Additives employed were: 2-amino-octanoic acid; arginine hydrochloride; ethyl vinyl ether; guanidine hydrochloride; hydroxyproline; mercuric acetate; monosodium glutamate; potassium chloride; sequestric acid; serine; α-tocopherol; urea; 2-amino-octanoic acid plus ascorbic acid, ethyl vinyl ether, hydroquinone, sequestric acid, tetrahydrofuran; decylamine plus acetic acid; glycine plus acetic acid, sequestric acid; serine plus ethyl vinyl ether, guanidine hydrochloride.

In control experiments hydrolysates of all but pure chloroform eluates showed fairly constant patterns of amino-acids (for typical patterns see refs. 11 and 14). In many of the additive experiments the patterns were little changed, and in the rest they were changed but not absent. Artefacts formed by several amino-acids, and by decylamine, were detected in amounts comparable with amounts of the usual lipoamino-acids present. For example, in experiments with 2-amino-octanoic acid, this acid appeared in hydrolysates of all except pure chloroform eluates; it became bound in *Drosophila* lipids if added at any stage before chromatography.

Antioxidant additives failed to prevent the appearance of lipoamino-acids or the formation of artefacts. 2-Amino-octanoic acid formed large amounts of artefact in presence of ethyl vinyl ether, suggesting that vinyl ether groups of the naturally occurring plasmalogens may be involved in artefact formation. Lipoamino-acids do not themselves appear to be plasmalogens, since treatment of whole horse blood lipid with 90 per cent acetic acid at 65° before chromatography did not significantly alter the amino-acid pattern in hydrolysates.

Neglecting the unlikely possibility that in the experiments described above additives failed to equilibrate with endogenous amino-acids, it is concluded that, while some lipoamino-acids are artefacts, others do exist *in vivo*.

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IS THERE A LEAKAGE OF ACETYLCHOLINE FROM POSTGANGLIONIC PARASYMPATHETIC NERVE ENDINGS?

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WHEN the chorda tympani is cut, a supersensitivity to chemical agents develops in the submaxillary gland, as first shown by Maevsky[1] and Fleming and MacIntosh[2]. A supersensitivity of similar type can be created by treating an animal for some weeks with ganglionic blocking compounds[3] or atropine[4]; since the sensitization is unspecific, secretory agents such as adrenaline can be used to show the effect of atropinization. These observations suggest that the gland cells acquire an increased sensitivity toward chemical stimuli when deprived of secretory impulses[5], and this hypothesis is supported by the finding that some supersensitivity can be produced by cutting afferent fibres of the reflex arc, the lingual or glossopharyngeal nerves[6]. As further evidence in this direction the fact may be quoted that the supersensitivity obtained after section of the chorda can be abolished by daily subcutaneous injections of secretory drugs such as pilocarpine or carbachol; even the responsiveness of the normally innervated gland can be lowered by this procedure[7].

A closer study of the effect of treatment with parasympatholytic agents reveals, however, that the transmitter, released by the nerve impulse, cannot be the only factor which determines the degree of sensitivity of the gland cells.

When atropine treatment is used to create supersensitivity, the doses of the drug have to be increased progressively since tolerance develops; if the initial small dose is given for a long period the sensitivity will first rise, and then decline again in spite of continued treatment (Emmelin, N., and MacIntosh, F. C., unpublished work, 1955). It is therefore difficult to compare the maximal level obtainable by atropinization with that brought about by section of the chorda. By using another parasympatholytic agent, piperidino-ethyl-diphenyl-acetamide (Hoechst 9980), it is possible to produce a marked supersensitivity without increasing the dose of the drug[8]. The supersensitivity obtained in such experiments is found to be much higher than that following section of the chorda[9]. This drug is very specific as a parasympatholytic agent; in the doses used it has, for example, no ganglionic blocking action and does not change the sensitivity to adrenaline in the acute experiment. It therefore seems reasonable to assume that it exerts its sensitizing action by virtue of its parasympatholytic properties. The supersensitivity caused by this drug should then in part be due to the fact that this agent, just as section of the chorda, deprives the gland of the action of acetylcholine liberated by secretory impulses from the central nervous system; but the sensitization above the chorda-level suggests that the drug in addition abolishes some action of a cholinergic mechanism which is left intact when the chorda is cut. In order to test the hypothesis that the supersensitivity caused by Hoechst 9980 is entirely due to loss of action of acetylcholine on the gland cells, some other parasympatholytic agents have been tried: methylscopolamine, ∫achesine and isopropamide (diphenylcarbamoyl - propyl - di*iso*propyl - methylammonium iodide). They all increase the sensitivity of the gland, all to the same level and to the level characteristic of treatment with Hoechst 9980 (unpublished work).

Section of the chorda tympani is a preganglionic denervation. It leaves the parasympathetic, cholinergic, postganglionic neurone intact. The obvious experiment would be to cut these fibres and investigate whether a supersensitivity ensues which is of the same size as that produced by treatment with parasympatholytic agents. Unfortunately, some of the synapses of the parasympathetic fibres are situated within the gland and not anatomically accessible. Many, however, are found close to the gland but still outside it, and by dissecting the nerve towards the hilum it is possible to produce a partial postganglionic parasympathetic denervation. A supersensitivity is found to develop which is not as high as that produced by treatment with parasympatholytic drugs but more pronounced than that following section of the chorda (unpublished work). Care has to be taken not to damage the postganglionic sympathetic fibres; this in itself would cause a supersensitivity which can be superimposed upon that due to section of the chorda[10]. In the parotid gland of the cat, it is feasible to cut all the postganglionic parasympathetic fibres by section of the auriculo-

temporal nerves. This operation produces a supersensitivity towards acetylcholine which is larger than that caused by preganglionic denervation[11]. It is, however, not possible to make comparisons between this sensitization and that produced by treatment with parasympatholytic agents because of lack of a suitable test drug; acetylcholine cannot be used since its secretory effect is abolished by such drugs. Adrenaline has to be given in large doses to produce a secretion from the parotid gland, and interfering vascular effects render quantitative comparisons impossible.

The question arises whether there is any indication of an activity in the postganglionic, parasympathetic neurone, apart from that due to impulses from the superior salivary nucleus, which results in a release of acetylcholine. There are two observations which could be quoted in support of the view that small doses of acetylcholine are continuously liberated from the endings of the postganglionic neurone, even when the chorda has been cut.

First, when the submaxillary gland is perfused with an eserinized solution, very small amounts of acetylcholine can be detected in samples of perfusate collected when the chorda has been cut acutely but is not stimulated[12]; this is the case even when the perfusion fluid contains curarine[13].

Secondly, eserine is known to cause a flow of saliva. This effect can be obtained when a small dose of eserine is injected intra-arterially near the gland[14] or through the duct towards the gland[15]. A very small dose, given through the duct, has no effect of its own but increases the secretory effect of injected acetylcholine. If the dose of eserine is raised, a flow of saliva is evoked even when no acetylcholine is administered. It is reasonable to connect this secretory action of eserine with an anticholinesterase effect. This is further supported by the fact that other cholinesterase inhibitors, like *TEPP* and paraoxon, cause a similar secretion. The secretory effect of these drugs is obtained after section of the chorda, and it is then as big as when the chorda is intact. If the secretion is due to acetylcholine, preserved by the esterase inhibitors, this acetylcholine has therefore not been released by impulses from the central nervous system. The secretory effect of eserine remains when the chorda fibres have been cut in advance and allowed to degenerate; after about a week or more it is even larger than for the normally innervated gland because of the sensitization of the gland cells.

Corresponding results are obtained in the parotid gland; but in this preparation it is possible to analyse further the secretory action of anticholinesterases[16]. When the postganglionic parasympathetic fibres have been cut and left to degenerate, eserine injected through the duct has only a very small secretory effect, in spite of the fact that the gland is even more sensitive to acetylcholine than after preganglionic section. Therefore, it seems legitimate to conclude that the main secretory action of eserine is connected with the appearance of acetylcholine originating from the endings of the postganglionic neurone. Such a release cannot be the result of impulses from the cell-body of this neurone, for the secretion is obtained when the postganglionic fibres have been severed acutely; nor is it likely, in such experiments, to be derived from impulses set up from the injured nerve stump, for the secretion is of the same size as that obtained from the contralateral normally innervated gland. A plausible explanation seems to be that there is a leakage of acetylcholine from the postganglionic parasympathetic endings similar to that which takes place at the somatomotor nerve terminals. It was, in fact, suggested already in 1945 by Feldberg[17] that there might be a more or less continuous release of small amounts of acetylcholine from the endings of cholinergic nerves in general. The amounts of acetylcholine released in this way should then be too small to cause any secretion; this would be the case even if the gland cells were sensitized by previous preganglionic denervation. On the other hand, if this acetylcholine should have anything to do with supersensitivity, it must in addition be assumed to exert some kind of action on the gland cells, the removal of which by postganglionic degenerative section or treatment with parasympatholytic agents manifests itself in supersensitivity. A higher level of supersensitivity after post- than after pre-ganglionic denervation is implied in Cannon's 'law of denervation'; leakage of transmitter at the postganglionic endings could provide an explanation for this phenomenon.

In the presence of an anticholinesterase the leaking acetylcholine must be assumed to accumulate to a concentration high enough to cause a flow of saliva. Such a concentration may be reached, under special conditions, even when no esterase inhibitor has been administered. One to three days after section of the postganglionic parasympathetic fibres of the parotid gland, a curious secretory activity can be found[18,19]. The flow is intermittent, occurring in bursts, and has been termed 'paroxysmal secretion'. Since it is abolished by atropine, it seems to be due to acetylcholine. It is, likewise, abolished by cocaine, injected through the salivary duct in a dose which does not change the responsiveness of the gland to injected acetylcholine; it is therefore likely that the acetylcholine responsible for the secretion is of nervous origin. It is reasonable to conclude that the acetylcholine is liberated from the endings of the degenerating nerve fibres. Since the flow persists when a local anæsthetic has been applied to the cut end of the nerve, the secretion cannot be initiated from that site. Acetylcholine seems to be released from the degenerating terminals in bursts in which the threshold concentration for secretion is temporarily surpassed.

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LETTERS TO THE EDITORS

RADIOPHYSICS

Global Hydromagnetic Wave Ducts in the Exosphere

In 1957, one of us (H.A.B.) considered the initiation of an earth-magnetic field experiment to detect hydromagnetic waves in the upper ionosphere and exosphere. He suggested the use of very large wire loops (of about 100 km.^{2} area) as pick-up systems for earth-magnetic field perturbations. A large loop has the advantage of a very high signal-to-noise ratio, since it effectively averages out local magnetic interference.

Acting on this suggestion, Berthold and Harris of this Laboratory installed two wire-loop stations (in Arizona and New Jersey), which were operated simultaneously during the summer of 1958. In addition to the advantages due to the high sensitivity of the loops, very accurate synchronization of the two stations was possible. This accuracy made it possible for Berthold and Harris to measure, for the first time, exact time differences in the arrival of sudden commencements at different parts of the world[1].

During the *Argus* experiments of 1958, Berthold and Harris recorded, at both stations, magnetic signals that originated from the high-altitude detonations. Both stations recorded two signals: a fast one, which arrived with a travel time of about four seconds; and a slower one, which arrived about four to five times later than the fast signal[2]. Similar signals were also observed by an earth-current station operated by the U.S. Army Signal Research and Development Laboratory in Maine under the supervision of G. Carp. In addition, the same two signals were recorded during the *Argus* experiments by two stations (Uppsala, Sweden, and Reykjavik, Iceland) operated under the supervision of H. Benioff of the California Institute of Technology, and by a geomagnetic station operated by the U.S. Air Force at the Azores[3]. Table 1, column 2, gives the observed times of arrival of the slow signal at these six stations, and Table 2, column 2, gives the same results for the faster signal.

Table 1. Slow Mode (Transverse Hydromagnetic Mode)

	T (Experimental) sec.	$K \cdot \int f(\varphi)$	P	T (Theoretical) sec.
Arizona ..	26·0	$16{\cdot}55 \times 10^{-26}$	$1{\cdot}57 \times 10^{26}$	24·8
Uppsala ..	15·7	$11{\cdot}55 \times 10^{-26}$	$1{\cdot}36 \times 10^{26}$	17·4
Reykjavik ..	17·8	$12{\cdot}45 \times 10^{-26}$	$1{\cdot}43 \times 10^{26}$	18·7
Maine	17·7	$11{\cdot}10 \times 10^{-26}$	$1{\cdot}59 \times 10^{26}$	16·7
New Jersey ..	18·3	$10{\cdot}90 \times 10^{-26}$	$1{\cdot}68 \times 10^{26}$	16·4
Azores ..	10·8	$8{\cdot}00 \times 10^{-26}$	$1{\cdot}35 \times 10^{26}$	12·0

$K = \sqrt{4\pi}/H_0a^3 = 4{\cdot}58 \times 10^{-26}$ [gauss^{-1}.cm.$^{-3}$]
$P = 1{\cdot}50 \times 10^{26}$
$P = R^4\sqrt{\rho}$ [cm.^{4} × (gm./cm.^{3})$^{1/2}$]

Table 2. Fast Mode (Longitudinal Hydromagnetic Mode)

	T (Experimental) sec.	$K \cdot \int f(\varphi)$	P	T (Theoretical) sec.
Arizona ..	5·0	$8{\cdot}11 \times 10^{-26}$	$6{\cdot}17 \times 10^{25}$	5·3
Uppsala ..	4·3	$7{\cdot}10 \times 10^{-26}$	$6{\cdot}06 \times 10^{25}$	4·6
Reykjavik ..	4·6	$7{\cdot}05 \times 10^{-26}$	$6{\cdot}53 \times 10^{25}$	4·6
Maine	4·5	$6{\cdot}96 \times 10^{-26}$	$6{\cdot}48 \times 10^{25}$	4·5
New Jersey ..	4·7	$6{\cdot}92 \times 10^{-26}$	$6{\cdot}80 \times 10^{25}$	4·5
Azores ..	4·2	$5{\cdot}95 \times 10^{-26}$	$7{\cdot}00 \times 10^{25}$	3·9

$K = \sqrt{4\pi}/H_0a^3 = 4{\cdot}58 \times 10^{-26}$ [gauss^{-1} . cm.$^{-3}$]
$P = 6{\cdot}52 \times 10^{25}$
$P = R^4\sqrt{\rho}$ [cm.^{4} × (gm./cm.^{3})$^{1/2}$]

Arguing that the observed signals were caused by the propagation of hydromagnetic waves due to the sudden displacement of earth-magnetic force lines by the highly diamagnetic fireball, we arrived at the following conclusions: (1) the hydromagnetic waves at high altitudes are essentially confined in ducts, which in first approximation are concentric shells about the earth; (2) within these ducts, the hydromagnetic waves travel essentially along great circle paths; (3) the propagation modes in these ducts are the well-known transverse and longitudinal hydromagnetic-wave modes.

If ds is a line element of a great circle path in the assumed ducts, the travel time between two points is given by:

$$T = \int ds/v$$

If we tentatively identify the slow signal with a generalized Alfvén wave, then $v = v_a\cos\theta$, where $v_a = H/\sqrt{(4\pi\rho)}$, and θ is the angle between the field lines and the direction of propagation. The fast signal should then be identified with the longitudinal hydromagnetic wave, for which $v = v_a$, since in the upper atmosphere v_a is about one thousand times as great as the speed of sound.

Thus, we obtain the following expression for the generalized Alfvén mode of propagation through the duct:

$$T = R^4\sqrt{\rho}\cdot\frac{\sqrt{4\pi}}{H_0a^3}\cdot\frac{1}{2\sin\varphi_0}\left[\ln\frac{\sin\varphi_0+\sin\varphi_2}{\sin\varphi_0-\sin\varphi_2}-\ln\frac{\sin\varphi_0+\sin\varphi_1}{\sin\varphi_0-\sin\varphi_1}\right]$$

where R is the distance of the duct from the Earth (centre to centre); ρ is the ion mass density in the duct; $H_0 = 0{\cdot}3$ gauss (surface magnetic field-strength of the Earth at the equator); $a = 6{,}371$ km. (radius of the Earth); φ_1 and φ_2 are the magnetic latitudes of the site of detonation and receiver stations, respectively; φ_0 is defined as the maximum magnetic latitude of the great circle path, that is, the most northerly point of such a great circle. This value can be readily obtained by elementary trigonometrical computation from the values of φ_1, λ_1, φ_2, and λ_2.

In similar fashion, the following expression is obtained for the longitudinal propagation mode:

$$T = R^4\sqrt{\rho}\cdot\frac{\sqrt{4\pi}}{H_0a^3}\int_{\varphi_1}^{\varphi_2}\frac{\cos\varphi\,d\varphi}{\sqrt{[(\cos^2\varphi-\cos^2\varphi_0)\,(1+3\sin^2\varphi)]}}$$

where the integral is an elliptical type. The values of this integral for the six stations are given in Table 2, column 3. In both expressions for T, we determined the value of the parameter $R^4\sqrt{\rho}$ that best fitted the experimental data. Table 1, column 5, gives the computed times of travel for the slow mode, and Table 2, column 5, gives the computed times for the fast mode.

The remarkable agreement between theoretical and experimental values, as evidenced by a comparison between columns 2 and 5 of Table 1, and between columns 2 and 5 of Table 2, seems to be a strong empirical indication of the existence of hydromagnetic wave ducts. The existence of such ducts would also explain better the observed intensity and sharpness of the signals received.

By the use of careful compilation of recent observations of ion density as a function of height in the upper atmosphere[4], and current estimates of the composition of the upper atmosphere, we computed the empirical function $R^4\sqrt{\rho}$ versus height, and found that our values for $R^4\sqrt{\rho}$ for the slow and fast modes corresponded to heights of 1,700 km. and 2,500 km. respectively, for the two ducts.

The refraction mechanism needed to bend most of the high-altitude hydromagnetic energy, produced by the detonation, into a very high-altitude duct has not yet been studied in great detail. Also, the correct mathematical relation between wave normal and propagation direction for our case of concentric ducts in an anisotropic medium has not yet been worked out. However, from the well-known formula for the radius of curvature of a beam being propagated in an inhomogenous medium, $r = v/dv/ds$ (where ds is a line element vertical to the direction of propagation), it follows (for longitudinal hydromagnetic waves) that the beam will be bent into a circular path around the Earth at a height where the ion density changes in proportion to R^{-3}. By using the empirical ion density versus height data, we find that this phenomenon occurs at a height about midway between 2,000 and 3,000 km., which independently supports the duct hypothesis.

A more detailed paper is being prepared for publication.

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GEOPHYSICS

Tektites and the Earth

The glassy tektites found strewn over the surface of the Earth have long been a scientific enigma. Many hypotheses have been produced to account for their origin and many writers seem to favour an origin beyond the atmosphere, in inter-planetary space. At this juncture it is well to consider several factors that argue against an extra-terrestrial origin.

The bulk constituents of tektites are the same as the constituents of certain sedimentary rocks[1], particularly sandy clays and shales; the twisted silica particles are almost certainly fused grains of sand; tektites are always found associated with sediments. These are arguments in support of the thesis that tektites result from a fusion of the sedimentary rocks of the Earth.

There are notable discrepancies in the percentage of minor constituents of tektites and sediments, but this may be attributed to leaching and weathering processes acting on the sediment before the tektite was formed. The minor constituents that are not subject to differential weathering all support a terrestrial origin. For example, the ratio of nickel to cobalt in tektites is 2·5, in close agreement with sediments[1], while meteorite material invariably shows a ratio between 6 and 60. The deuterium/hydrogen ratio[1] is 0·015 mole per cent, which agrees with the ratio found in the natural waters of the Earth and is considerably higher than the value accepted for non-terrestrial objects. The ratio between strontium-87 in tektites to the total is 0·0702. This value does not correspond to the values for stony meteorites, but agrees closely with the strontium-87 content of sea water and marine sediments[1]. A measurement of the ratio of lead isotopes again shows agreement with terrestrial rocks, and the uranium-thorium-lead balance[1] is more characteristic of Cainozoic sediments than any other material that the astronomer or geologist would expect to find in the entire solar system.

One of the most significant ratios for meteorites is that of iron to nickel, which is usually close to 20 and seldom, if ever, greater than 100. Unfortunately the ratio of iron to nickel on the Earth varies over wide limits for the various types of rock, and is probably affected by differential leaching and weathering in sedimentary deposits. Despite these uncertainties it is of interest to consider the iron-nickel ratio. Tektites show a ratio between 500 and 3,000[1,2], which is considerably higher than that of meteorites and corresponds to the average sediment. Collisions in space are responsible for the dispersal of nickel-iron fragments in stony meteorites and even the surface of the Moon must be contaminated with the 20 : 1 iron-nickel ratio. Thus one would have expected a higher percentage of nickel if tektites were connected in any way with an extra-terrestrial object, whether it be the Moon, an asteroid or meteorite.

Radioactive dating shows that the age of tektites agrees fairly closely with the age of the sedimentary rocks in the tektite area[1]. That is to say, the tektites were probably molten fragments at some instant during the deposition of the sediments. Cosmic-ray bombardment produces helium, neon, argon and other spallation isotopes in the meteorites, which give a measure of the duration of exposure of the specimen in space. So far no measurement has been able to show that the exposure time of tektites is different from zero, and we must conclude that after the melting process tektites were screened from cosmic rays, presumably by the atmosphere of the earth.

These facts offer convincing evidence that tektites were formed on the Earth's surface from sandy sediments by the momentary application of intense and localized heat. I consider that it is more prudent to assume as a working hypothesis that tektites are earthy objects and consider extra-terrestrial origins only when the hypothesis fails.

The suggestion that tektites were produced by the collision of meteorites or comets with the Earth is consistent with a terrestrial origin. The energy of impact would be sufficient to melt the country rock within and around the crater. Impact theories do not adequately account for the widespread nature of tektite fields such as those in Australia and the East

Indies. Furthermore, the original craters have never been found, although we recognize other craters several million years old, nor do tektites contain iron spherules as are found in the glass near recognized meteor craters.

Fusion by lightning has been discounted by many authors[1] because lightning usually produces a long tubular fulgurite whereas tektites are solid and sometimes show pronounced stream-lining or 'flight form'. Fulgurites formed in sand are certainly tubular, but the action of lightning on other sediments, such as a sandy clay, is not known. Recently clay fulgurites, formed in Kingston, Ontario, were brought to my attention by Wallace H. Robb. The objects were partially fused ellipsoids ranging in size from an inch to several inches. We hope to make further studies on these objects. It is perhaps possible for the buttons with flanges, dumbbells and other shapes to be formed by fusion within the ground. It is not correct to assume that these shapes can only be produced by the melting of a tektite during supersonic passage through the air. The usual flight form of meteorites is an irregular cone, the material being blown away as fast as it melts. Meteorites never show flanged buttons, dumbbells, etc.

At Boston University we have commenced a study of tektites, with the support of a N.A.S.A. research grant, and we are considering the hypothesis that tektites are produced by lightning acting on sandy sediments. Lightning offers a ready explanation for the chemical and isotope composition of tektites, their poor annealing and fused silica particles, their age and their widespread yet infrequent occurrence. Artificial tektites have already been produced[2] by the action of a high-energy electron beam on the sandy clay found, on a recent field trip[4], to be associated with the tektites in Texas. Further experiments will continue in an attempt to reproduce the 'flight forms' by fusion, abrasion and etching.

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[See also p. 291 of this issue.—EDITORS.]

PHYSICS

Time of Residence of Radioactive Debris in the Stratosphere

THE world-wide distribution of strontium-90 from nuclear weapons tests has been extensively studied, especially by the University of Chicago Sunshine Project and others. It has been established that the cumulative deposition has a strong peak in the middle latitudes of the northern hemisphere, and the rate of deposition has repeatedly shown a maximum in the spring.

Libby[1] assumed originally that the debris injected into the stratosphere has a mean time of residence of 10 ± 5 yr. and will be distributed rather uniformly over the globe. He ascribes the peak in the middle latitudes and the increase in deposition in spring to the tropospheric part of the debris.

Based on extensive measurements, Stewart *et al.*[2] have maintained that at places remote from testing areas nearly all strontium fallout is of stratospheric origin. They propose a selective downward mixing of stratospheric debris at middle latitudes, which could explain both the peak and the increase in the spring. Martell[3], however, submits an alternative view; namely, that these phenomena are due largely to recent tests ofintermediate- and high-yield weapons which have injected debris into lower and intermediate levels of the stratosphere, debris which has a relatively short time of residence in the stratosphere.

Libby and others[4] now also evaluate the time of residence for stratospheric debris at middle latitudes as considerably shorter, perhaps 2–4 yr.

We have for some time made determinations of the ratio, r, of the amounts of strontium-89 and strontium-90 on monthly samples of precipitation from a number of stations in Denmark, including the Faeroes and Greenland. The location of the stations, given to the nearest degree, are as follows:

		Longitude	Latitude
B	Oestermarie, Bornholm	15 E.	55 N.
C	Copenhagen, Zeeland	13 E.	56 N.
T	Thorshavn, The Faeroes	7 W.	62 N.
G	Groennedal, Greenland	48 W.	61 N.
S	Soendre Stroemfjord, Greenland	50 W.	67 N.

Our results are given in Fig. 1. The accuracy of the measurements varies with the strength of the samples as shown.

It can be seen that the ratio r in August and September 1958 is around 15. As part of the fallout may be tropospheric, $r = 15$ is an upper value for debris from the stratosphere at that time. The ratio increases in October for some (the northernmost) stations to a little less than 40. From February 1959 it again decreases for all stations. The two dotted lines are drawn with a slope corresponding to the decay of strontium-89.

We believe that the two results for Copenhagen in October and November indicate that very little dust from explosions in October reach Copenhagen before December.

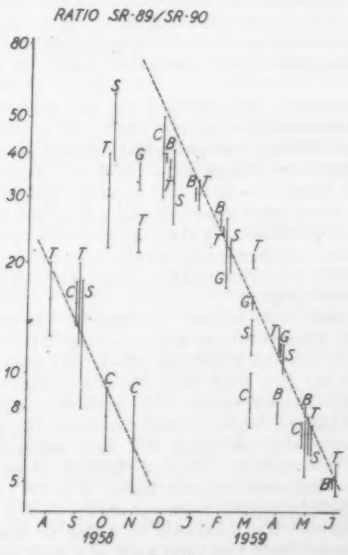

Fig. 1

The results for the period January–June follow the slope of the lines remarkably well. Combined with the cessation of weapon tests around November 1, this seems to show that for a period of five–six months starting two–three months after the last injections in the stratosphere, and for 5 stations distributed nearly along a line 2000 miles long, the composition of the fallout corrected for the decay is very uniform.

Since the time of residence in the troposphere is much shorter and stratospheric debris from older tests would give a lower value of r, it is concluded that a considerable part of the fallout in the spring of 1959 over a large area was stratospheric, but originated from the tests in northern latitudes in the autumn of 1958.

From the increase in r as given by the vertical distance between the two lines in Fig. 1, it is inferred that 40–50 per cent of the strontium-90 fallout in the period February–June comes from these tests.

These measurements are part of work carried out for the Danish Defence Research Board, and I thank the Board for permission to publish these results.

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Measurement of Ion Beam Currents using a Hall Effect Magnetometer

Ion and electron beam currents are normally measured in terms of either a collected current or the beam power dissipated in a calorimeter. Direct-current collection is complicated by secondary emission at the collector surface, and calorimeters are difficult to use when the beam power is low. For both methods the beam must be interrupted in order to measure the current. The technique described here avoids these disadvantages by measuring the magnetic field associated with the particle beam.

The magnetic field-strength in the vicinity of a beam of charged particles is given by $H = i/5r$, where i is the net current in amperes and r the radial distance in cm. from the beam axis. If $i = 1$ m.amp. and $r = 1$ cm. then $H = 2 \times 10^{-4}$ oersted. Fields of this intensity have been detected using the Hall effect in indium antimonide[1]. If the magnetic field generated by the beam is concentrated on the indium antimonide crystal then currents smaller than one milliampere may be detected.

A 'Mumetal' rod 1·3 cm. in diameter was bent into a 6·0-cm. diameter ring and annealed. An indium antimonide crystal 0·025 cm. thick was placed in a 0·040-cm. air gap built into the ring. This indium antimonide magnetometer was used in conjunction with a transistor oscillator and amplifier. In order to demonstrate the principle the ring assembly was mounted in a vacuum chamber through which passed a 1·0-cm. diameter electron beam of 1 keV. energy. Two current balancing loops, as shown in Fig. 1, were used to cancel the magnetic field due to the beam. The magnetometer was then used as a null indicating

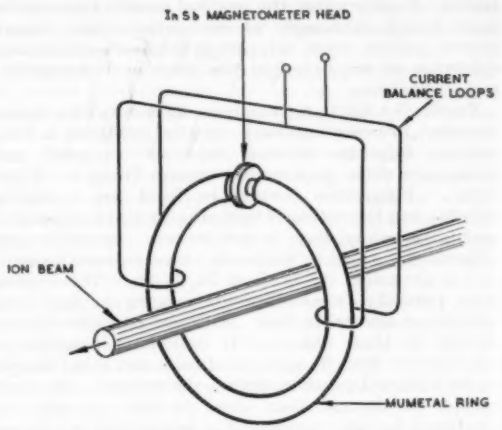

Fig. 1. Schematic diagram of magnetometer ring assembly

device and at the null point the beam current equalled the current through the balance loops.

Two methods of applying the magnetometer were investigated. In the first method a steady electron beam was directed through the ring with the crystal excited by a 1 kc./s. alternating current. A balance was obtained by adjusting the current through the balance loops until the value of the 1 kc./s. Hall voltage equalled that for zero beam current. The smallest beam current detectable above the noise-level was about 10 μamp. Although a balance was easily obtained for larger currents the accuracy of the device was mainly limited by the zero drift caused by small changes in temperature of the crystal unit. Displacement of the beam from the centre of the ring did not appear to alter the balance condition.

In the second method the crystal was excited by a steady direct current while the beam was modulated at 1 kc./s. Only the alternating component of the Hall voltage was fed into the amplifier. In this case zero drift was virtually eliminated and the smallest detectable current was slightly less than 10 μamp.

The technique offers a new experimental approach to certain problems involving ion beam current measurement. Ionic processes may be studied without the complications associated with conventional detectors.

We are grateful to Miss P. Dunmow and Mr. N. A. C. Thompson for their help in the experiments. Permission to publish has been granted by the Admiralty.

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Cavity Formation in Simple Pipes due to Rupture of the Water Column

When the water-hammer surges that accompany rapid velocity changes in pipes are of such a kind as to produce a fall to vapour pressure, cavities are formed. Theory and analysis at present available rest largely on empirical evidence of changes in pressure alone. In the case of rupture, a recent publi-

cation appears to support methods of analysis already in use[1], and suggests cavity formation would take place at the control (gate) end of a straight horizontal pipe.

Studies of the changes in pressure at the centre of a 60-ft. horizontal length of 1-in. copper tube as well as near the control end suggest a new but logical development of the normal theory. The usual principle of the independence of the direct and reflected waves expressed in the basic wave equations can no longer be tacitly accepted, and any such waves which may meet along a pipe must be algebraically summed to ascertain whether the resultant pressure tends to yield a value less than or equal to vapour pressure. It is believed that such a tendency will be invariably found to exist during all rupture periods that occur with the exception of the first. Because pressures less than vapour pressure cannot be tolerated, additional cavity formation is necessary to ensure compatibility of pressure and velocity changes.

Fig. 1 illustrates the result of the theoretical analysis proposed for a specific example. Additional cavity formation occurs at point D, due to the simultaneous arrival at this point of a normal reflected wave and a new direct wave occasioned by the resurge of the water column at A at the end of the first rupture phase.

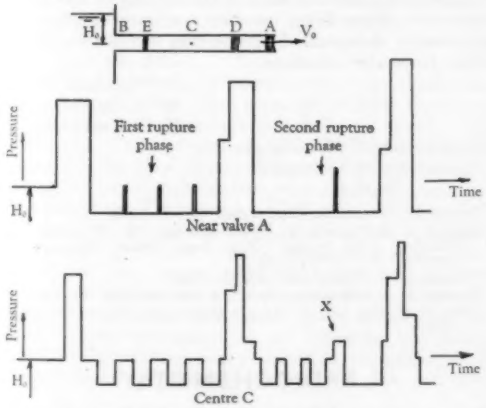

Fig. 1. Graphical solution for rupture

The cavity at D in the example shown disappears before the normal cavity at A, and evidence of this is seen in the positive pressure rise (marked X) at C during the second rupture phase. In the case shown further peculiarities of the pressure variations are due to the formation of a complementary cavity at E.

Pressure records support the theory, and also observations of a transparent pipe with photographic aid confirm the formation of the cavity as predicted.

It is considered that analytical and experimental investigations of this cavity formation will enable: (a) The phenomenon of rupture of the water column to be explained. (b) The growth and decay of cavities under the action of estimable forces to be described, including the effects of entrained air. (c) The assumed mathematical relationship between pressure and velocity changes to be studied, in view of the fact that the moving cavity serves as an index of water velocities that exist during certain phases.

Work which has been in progress for some time is proceeding along these lines.

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METALLURGY

A Model of Metal Fatigue

The important part played by the disturbance of the surface of a fatigue specimen by to-and-fro slip movement in the slip bands has been emphasized by much recent work, especially that of W. A. Wood[1]. If the slip in alternate cycles were exactly reversed in the same atomic planes, then there would be no surface disturbance. On the other hand, extrusions or intrusions will be formed if by some mechanism such as that proposed by Mott[2] the dislocations which have moved in one direction along a certain path return by a different one, so as to go round a closed circuit in the same sense at each cycle. There is another possibility: that the dislocations make their return journeys along paths which are shifted in a random fashion with respect to the previous paths. If the shifts are comparable with the width of the slip band, this is equivalent to assuming that the slip is distributed at random at each half-cycle among the available slip planes, and that the distribution of slip at each half-cycle is independent of the distribution of slip in previous cycles.

This random distribution of slip will roughen the surface, each part of which is engaged in a random walk along the slip vector. This roughening will lead to a re-distribution of the stress, so that in later cycles the slip will tend to be concentrated in the valleys already formed on the surface. The subsequent surface movements will make some of the valleys less deep, but some will become deeper, and on these the slip will be even more concentrated. I have set up a rough model of this process in mathematical form, in order to see whether valleys deep enough to lead to cracks could be formed in a reasonable number of fatigue cycles.

These calculations will be published elsewhere. The main result is that after t cycles the fraction f of the valleys of width w which have depths between z and $z + \mathrm{d}z$ is given by:

$$f = F \exp - [\{\sqrt{(z + w)} - \sqrt{w}\}^2 / b\, \varepsilon\, t]$$

where F is a slowly varying function of z, w and t. The slip vector b and the plastic strain ε in the slip band are taken as constant. The distribution consists of a Gaussian peak near $z = 0$ and an exponential tail at large z governed by the factor $\exp - z/b\varepsilon t$.

If we suppose that at some large value of z, say Z, the valleys become deep enough to form cracks, we can regard $Z/b\varepsilon$ as a time constant T controlling the rate at which valleys attain the depth Z, and therefore the life-time of the specimen. The first valleys will attain a depth Z after approximately $T/15$ cycles.

It remains to make a rough estimate of the numerical magnitudes involved. We suppose that a valley with a depth ten times its width can be regarded as an

incipient crack. Thus $Z \sim 10w$, giving $T \sim 10w/b\varepsilon$. If we insert as typical values, $w = 10^{-4}$ cm., $b = 5 \times 10^{-7}$ cm. (corresponding to the passage of a group of about 20 dislocations at each slip), and $\varepsilon = 10^{-3}$, we get for T about 2×10^{6} cycles. The first incipient cracks will appear at about $1{\cdot}3 \times 10^{5}$ cycles. The satisfactory agreement with the order of magnitude found in practice for life-times suggests that it would be worth while to investigate a more realistic model of this type.

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Oxidation of Titanium–Iron Alloys

Above the transition temperature, the oxidation of titanium proceeds by a layer of titanium oxide (TiO_2) growing outwards by the migration of anion vacancies. Concurrently, a solid solution layer of alpha-titanium containing dissolved oxygen grows inwards. The alpha-layer is fairly uniform in thickness over the surface of the specimen. Microhardness measurements show a very marked hardness gradient across it, indicating an oxygen concentration gradient and suggesting that the rate of growth of the alpha-layer is controlled by the diffusion inwards of interstitial oxygen.

It has been found that the addition of small amounts of iron to titanium, while not markedly affecting the overall rate, modifies the structure of the alpha-layer. Alpha-plates form rapidly along some of the beta grain boundaries, plates about 0·5 mm. long forming in about 15 min. at 875°C. Later, the alpha-plates thicken slowly, and a two-phase alpha/beta layer of roughly uniform thickness is formed around the specimen and grows inwards. There is no hardness gradient within the alpha-plates, suggesting that oxygen diffusion is not the controlling step in their formation.

Fig. 1. Titanium–3 per cent iron oxidized 90 min. at 867° C. (×80)

However, when a specimen containing 3 per cent iron is quenched after oxidation, only the beta-phase remote from the alpha-plates is transformed martensitically, as is shown in Fig. 1. Duwez[1] has shown that above 4 per cent iron the martensite start temperature is less than room temperature and beta-phase is retained. From this it is deduced that the iron content in the beta-phase adjacent to the alpha-plates is greater than 4 per cent. Alpha titanium does not dissolve iron to an appreciable extent[2] and it would appear that the iron ejected from the advancing alpha-plates raises the concentration of iron significantly. It is not possible to determine the variations in the iron content of the beta-phase by micro-hardness measurements, because in the neighbourhood of 4 per cent iron the hardness of quenched titanium varies very little with iron content[3]. The oxygen content, as indicated by the hardness of the alpha-phase[4], is about 2 per cent by weight at 850°C. and 2·5–3 at 900°C. which, if local equilibrium is assumed, indicates an iron content of about 6–7 per cent by weight[5].

It is concluded, therefore, that there is an iron gradient in the beta-phase from the original 3 per cent at the centre of the grains remote from the alpha-plates to about 6 per cent by weight at an alpha/beta interface. The rate of thickening of the alpha-plates, and the rate of thickening of the alpha/beta two-phase layer at the specimen surface, are apparently determined by the rate of diffusion of iron away from the interface.

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RADIOCHEMISTRY

Preparation of Radiochemically Pure Tritium Nuclear-labelled Morphine

Morphine has been labelled with carbon-14 in the N-methyl position by chemical synthesis[1,2] and randomly by biosynthesis[3,4]. Because of the metabolic lability of the N-methyl group and the need for considerable quantities of a product of high specific activity, studies on the preparation of tritium-nuclear labelled morphine were initiated. The Wilzbach procedure[5] of exposure to tritium gas was utilized for the introduction of the tracer element into the morphine molecule. The preparation of biologically stable tritium-labelled morphine by this technique had been described previously[6], and the radiochemical purity of the product was reported. We have found, however, that the exposure to tritium gas of morphine base or morphine sulphate in finely divided form, and subsequent purification to remove the labile tritium and radio-decomposition products, uniformly gave a seemingly pure crystalline morphine which showed two distinct areas of radioactivity when paper chromatograms were developed using correctly buffered paper

and the proper solvent mixtures. One area contained a greater proportion of radioactivity (approximately 65–70 per cent of total) but negligible mass of material (not detectable by spray reagents or by visualization with ultra-violet light, less than 1 per cent). Similarly when normorphine base or its hydrochloride was tritiated in the same manner and purified, the morphine synthesized from it by the method of Andersen and Woods[1] showed the same trace radio contaminant. When paper chromatograms were prepared from the above tritium-labelled 'morphine' using non-buffered paper and several solvent mixtures, that is, *n*-butanol, formic acid, and water (12 : 1 : 7 v/v); 2·3 per cent *p*-toluene sulphonic acid in *n*-butanol and water (100 : 14-v/v), etc., all the radioactivity was located in one area. This area was the alkaloidal spot as localized by spraying with iodoplatinate alkaloidal reagent or by visualization of the shadow with ultra-violet light. The R_F values were identical with parallel non-labelled morphine.

Paper chromatograms obtained with paper buffered at *p*H values varying between 5·6 to 8·0 (Table 1) and the solvent systems of *tert*-amyl alcohol and water or *tert*-amyl alcohol, *n*-butyl ether and water (80 : 7 : 13 v/v)[7], effected a separation of the radioactive impurity from morphine. By the use of co-chromatography with non-radioactive dihydromorphine, the paper chromatograms gave presumptive evidence that the radiochemical impurity in the tritium-labelled 'morphine' was tritium-labelled dihydromorphine. Separation of the two areas of radioactivity was not possible when the paper was buffered at higher *p*H values, namely, 9·25.

Crude tritium-labelled normorphine hydrochloride was converted to free base, crystallized from methanol and the product reprecipitated from an acetic acid solution by dilute ammonia. (The exposure was performed by the New England Nuclear Corp., Boston, Mass.) The normorphine was converted to morphine by the method of Andersen and Woods[1]. The specific activity (128 μc./mgm.) of the resulting morphine did not change on three serial crystallizations. The amount of the trace radiocontaminant remained unchanged or was only partially removed by vacuum sublimation of the 'chemically pure morphine', or fractional crystallization of its diacetyl derivative followed by the conversion of acetylated product back to morphine. Attempts to remove the tritium-labelled dihydromorphine with basic or acid-washed alumina columns, or cellulose columns, were not successful.

Table 1. TYPICAL R_F* VALUES OF COLD AND RADIOACTIVE MORPHINE AND DIHYDROMORPHINE CALCULATED FROM CHROMATOGRAMS OBTAINED WITH WHATMAN NO. 1 PAPER

Compound	*p*H of buffer solution applied to paper				
	5·7[a]	7·0[b]	7·2[b]	7·9[b]	9·25[c]
Morphine	0·56	0·28	0·37	0·66	0·82
Tritium-labelled morphine	0·55	0·29	0·36	0·65	
Dihydromorphine (non-labelled)	0·41	0·15	0·18	0·54	0·82
Tritium-labelled 'dihydromorphine'	0·40	0·17	0·15	0·56	
Mixture of known non-labelled dihydromorphine and eluted area of tritium labelled 'dihydromorphine'	0·40				

The paper strips were dipped in the 0·2 *M* buffer solution, allowed to drip dry in a hood for 1–2 hr., placed in a glass cylinder containing the solvent mixture for 12–14 hr. (overnight) and the chromatogram then developed with the selected solvent mixture. The scanning of dried chromatogram strips was done by eluting 1-cm. strips with absolute methanol, evaporation in counting vials and subsequent counting in a Packard Tricarb liquid scintillation spectrometer.

* The values are not meant to be absolute and may vary somewhat with temperature, nature of buffer used or extent of saturation of paper in chamber. Non-labelled compounds were run as references in all experiments. The temperature was 23·5° ± 0·5° C. The values for radioactive compounds were calculated from the centre of maximum radioactivity.

[a] *Tert*-amyl alcohol, saturated with 1 : 1 (v/v), 0·2 *M* acetate buffer (this buffer was also used to saturate the paper).
[b] *Tert*-amyl alcohol, *n*-butyl ether and water (80 : 7 : 13 v/v) and paper buffered with 0·2 *M* phosphate buffer.
[c] 0·2 *M* borate buffer; solvent mixture as in footnote *b*.

Radiochemically pure morphine was separated by descending chromatography on Whatman 3 *MM* paper (5 mgm./large sheet) buffered with 0·2 *M* phosphate buffer, *p*H 7·0, and developed with a *tert*-amyl alcohol, *n*-butyl ether and water (80 : 7 : 13) system. The morphine band located under ultra-violet light was extracted with absolute methanol in a Soxhlet extractor.

The methanol extract was mixed with cold carrier and evaporated to dryness under nitrogen and reduced pressure. The morphine was separated from this residue and passed over a small neutral activated alumina[8] column to give pure morphine base. The specific activity was about 5–10 μc./mgm. with a one to three isotope dilution with cold carrier. The same procedure using cold dihydromorphine as carrier gave radiochemically pure dihydromorphine.

Morphine exposed to tritium by the Wilzbach procedure shows addition at the 7 : 8 double bond giving the dihydro-radiocontaminant having a negligible amount of total mass but a greater fraction of total radioactivity. Since this material is not biologically identical with morphine, it is essential to remove the former from morphine to be studied biologically. A detailed report will appear elsewhere.

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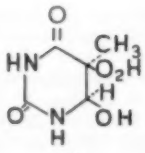
RADIATION CHEMISTRY

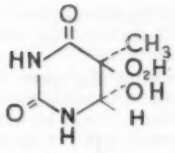
Organic Hydroxy-Hydroperoxides: a Class of Hydroperoxides formed under the Influence of Ionizing Radiations

IN continuation of our previous work on nucleic acids and related compounds[1], it has now been found that many substances, when irradiated with, for example, γ-rays, X-rays, in aqueous solution in the presence of oxygen, are capable of forming hydroxy-hydroperoxy compounds, many of which were previously unknown.

Compounds belonging to this class which may be of interest from a biological point of view are the hydroxy-hydroperoxides, which are formed from different pyrimidine bases, for example, thymine and uracil. Among these, the thymine hydroxy-hydroperoxide is rather stable and can be isolated from the irradiated solutions. In support of an earlier suggestion[2], it has now been found that the peroxides produced by radiation are the (*cis*) and (*trans*)-4-hydroxy-5-hydroperoxide (I) and (II):

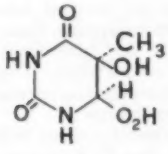


(I)

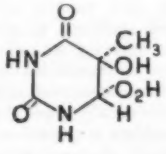


(II)

It is of interest to note that these compounds are not decomposed by catalase. The isomeric 5-hydroxy-4-hydroperoxide should also be possible in a *cis* and *trans* form, so that in addition to (I) and (II) the following compounds may exist :



(III)



(IV)

All these compounds have in fact now been made from thymine by chemical methods by Dr. B. Ekert[2] at the Pasteur Institute. They can be separated, for example, by paper chromatography by means of the solvent system *n*-propanol (80 parts), *N* hydrochloric acid (15 parts) on Whatman No. 1 paper and can also be characterized in this way.

Another convenient way of characterizing these different hydroperoxides was found by measuring the rate constants for the liberation of iodine in the reaction of the hydroperoxide with iodide in aqueous solution. The second-order rate constants for the different hydroxy-hydroperoxides from thymine are given in Table 1 :

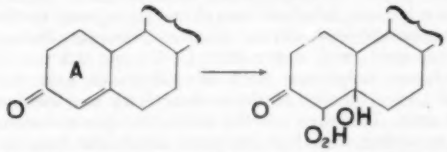
Table 1. Rate Constants for the Reaction of Different Hydroperoxides with Iodide at *p*H 4·0 and 25° C. (Measurements carried out by Mr. J. F. Ward)

Compound	Rate constant (l. mol.$^{-1}$ sec.$^{-1}$)
Hydroperoxide by radiation from thymine	1·59
4-Hydroxy,5-hydroperoxy thymine (I, II)	1·60
4-Hydroperoxy,5-hydroxy thymine (III, IV)	7·53
Hydrogen peroxide	$1{\cdot}13 \times 10^{-3}$

The rate constant for hydrogen peroxide, measured under the same conditions, has also been included : a comparison of the values shows that the thymine hydroperoxides are very much more reactive than hydrogen peroxide. The rate constant of the compound produced by radiation is practically identical with that of the 4-hydroxy,5-hydroperoxy isomer. The 4-hydroperoxy,5-hydroxy compounds (III, IV) are, under the conditions in question, nearly five times more reactive than the isomer with the hydroperoxy group in the 5-position. On the other hand, we could not detect any appreciable differences between the corresponding *cis* and *trans* isomers. The reason for this may be that either the differences are too small, or in solution a relatively rapid *cis–trans* isomerization takes place.

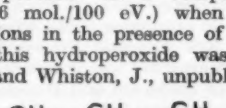
Uracil also forms a corresponding hydroxy-hydroperoxide which is of comparable stability ; on the other hand, the hydroperoxide from cytosine appears to be much less stable in solution.

Another new group of hydroxy-hydroperoxides has been obtained from various steroids by means of ionizing radiations[4]. Peroxides derived from steroids have recently again attracted some interest as possible sources of the carcinogenic effects of such substances. The compounds described by Fieser *et al.*[5] and Schenck *et al.*[6] are ordinary hydroperoxides. The study of the action of ionizing radiation on aqueous solutions of, for example, deoxycorticosterone, cortisone and related steroids has shown that these and similar steroids are capable of forming hydroxy-hydroperoxides. Although these compounds have not yet been isolated in a pure form, their chemical and spectroscopic behaviour shows clearly that the OH and HO_2 attach themselves to the double bond of ring *A*, namely :



and/or the corresponding isomer with the OH and HO_2 groups exchanged, which may also be formed to a lesser or greater extent. There are, of course, again two possible stereoisomers for each structure. This type of structure is supported also by the disappearance of the ultra-violet absorption associated with the chromophoric α,β unsaturated ketone grouping in ring *A*. In the case of deoxycorticosterone, which has been studied in some detail, it has been found that two different hydroperoxides are formed in the ratio of approximately 2 : 1. In addition to the hydroperoxide on ring *A*, a second one, which is associated with the side-chain, is also formed, as was shown by a corresponding decrease in the tetrazolium blue reaction.

With regard to some simpler olefinic molecules, it may be of interest to note that propylene forms a hydroxy-hydroperoxide in a comparatively high yield ($G = 2{\cdot}6$ mol./100 eV.) when irradiated in aqueous solutions in the presence of oxygen. The structure of this hydroperoxide was found to be (Clay, P. G., and Whiston, J., unpublished results) :



It can thus be stated that, in general, the action of ionizing radiations on ethylenic systems in the presence of oxygen can give rise to more or less stable hydroxy-hydroperoxides. These compounds may be of considerable biological interest, and bearing in mind the important and unique role of oxygen in connexion with the radiosensitivity of living cells, further study of the biological effects of such compounds is desirable.

We thank Dr. B. Ekert for supplying us with samples of the synthetic thymine hydroperoxides and the Rockefeller Foundation for financial support.

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Electron Paramagnetic Resonance Observations on Irradiated Thiols and Disulphides

THE unique role of sulphur in radiation damage is amply borne out by recent electron paramagnetic resonance work[1–3]. In a current investigation on the mechanism of action of the sulphur-containing radioprotective compounds, quantitative electron paramagnetic resonance studies have been carried out on a series of irradiated thiols and disulphides.

The substances were irradiated in the polycrystalline state at room temperature with conventional X-rays (220 keV.). Irradiations were carried out in vacuum as well as in air. In most cases, the radiation doses were less than 5×10^5 r. The spectrometer used operates at a frequency of 9,200 Mc. per sec. The spectra obtained represent the first derivative of the actual absorption curves. Quantitative results were obtained by double integration and comparison with measurements on standard systems. The more significant observations may be summarized as follows:

Dose-effect relationships. The qualitative electron paramagnetic resonance spectra exhibited in some instances a continuous and distinct change with increasing radiation dose, even in the comparatively low dose-range used (Fig. 1). Moreover, for all compounds studied the number of resonance centres detected did not increase linearly with the radiation dose, as exemplified in Fig. 2. This dependence of the yield on the radiation dose may in part explain the large discrepancies found in the literature concerning the number of electron volts needed to produce free radicals[4–6].

Time effects. The number of detectable paramagnetic resonance centres showed an unexpected relationship to the time elapsed after the irradiation. For some compounds (for example, cystine, penicillamine) the number of free radicals N seemed to decay in vacuum according to the general equation: $N = a \ln(t) + b$, where t is the time and the parameters a and b vary with the substances. In the case of other compounds (for example, homocystine, cystamine), the number of resonance centres increased by approximately 10–20 per cent during the first hours after the irradiation. In these instances the peak was reached in 3–4 hr. and the number then decreased, the decay obeying the general formula stated above. The quantitative changes were associated with qualitative changes in the electron paramagnetic resonance spectra in the case of homocystine and cystamine. The spectra of cystine and penicillamine, on the other hand, were constant with time.

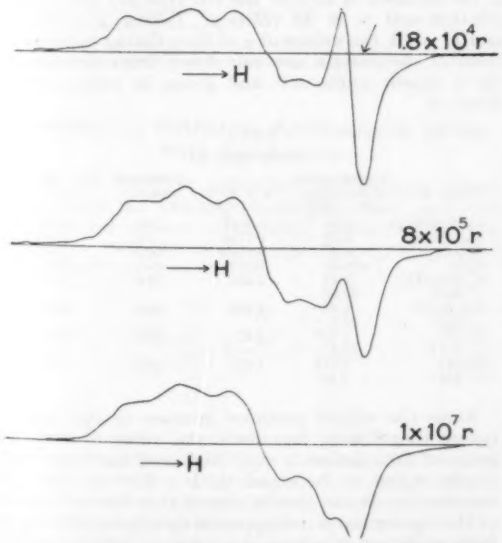

Fig. 1. The effect of the radiation dose on the qualitative electron paramagnetic resonance spectra of cystamine dihydrochloride irradiated in vacuum. The measurements were carried out 30 min. after the conclusion of the exposure. The arrow indicates the position of g-factor of free electron spin. The total width of the spectrum is about 150 gauss

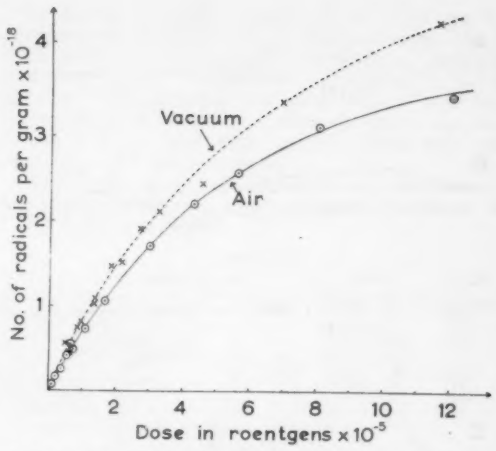

Fig. 2. The relationship between the radiation dose and the number of paramagnetic resonance centres induced in cystamine dihydrochloride. From the curves, 'initial yields' (calculated from the straight line portion of the curves) were found to be 7 and 8 eV. per induced spin in vacuum and in air, respectively

The observed increase in the number of resonance centres in the case of homocystine and cystamine suggests the possibility that several types of free radicals are initially formed, one of which may decay with the formation of two or more other types. The concurrent qualitative spectral changes may be taken to support this view.

Structural effects. In contrast with previous views[1], structural factors were found to exert a pronounced effect on the qualitative electron paramagnetic resonance spectra of thiols and disulphides (Fig. 3). Whereas cysteamine, cysteine, and cystine gave closely similar resonance patterns, marked differences were obtained when the carbon chain was extended (for example, 1-mercapto-7-aminoheptane, homocystine) or branched (for example, penicillamine). Substitutions in the amino group (N,N′-tetramethylcystamine, N-morpholyl-cysteamine, N-piperidylcysteamine) likewise resulted in pronounced changes in the resonance patterns. In general, these chemical alterations introduced a hyperfine structure in the high-field part of the spectra.

The above-mentioned findings bear out the fact that electron paramagnetic resonance measurements are influenced by several parameters which must be clearly defined in qualitative as well as in quantitative electron paramagnetic resonance studies. They also demonstrate that the chemistry of the radiation of thiols and disulphides in the dry state is more com-

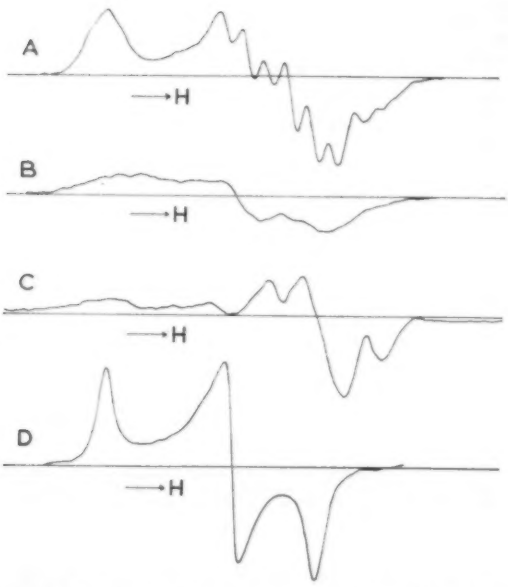

Fig. 3. Examples of variations in the qualitative electron paramagnetic resonance spectra with variations in the chemical structure. Dose, 5×10^5 r. Otherwise, conditions as in Fig. 1. *A*, N,N'-tetramethylcystamine dihydrochloride; *B*, *l*-cysteine hydrochloride; *C*, reduced glutathione; *D*, *d*-penicillamine. In the course of 2–3 days, the spectrum of glutathione changed to resemble closely that of cysteine

plex than previously recognized. The distinct differences observed between the spectra of various thiols and disulphides open up the possibility of studying the chemical properties of irradiated mixed disulphides, a question which is of interest from a radiation protection point of view[7,8]. Such studies are in progress.

The details of this work will be published elsewhere.

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CHEMISTRY

Experimental Test of Verwey and Overbeek's Theory

Ghosh and Rakshit[1] have measured the ζ-potentials of a titanic oxide sol in the presence of equicoagulating concentrations of electrolytes. An attempt has been made here to calculate the ζ-potentials of the colloid particles of a titanic oxide sol at flocculating concentrations of electrolytes from an analysis of the nature of the curves of total potential energy of interaction of two spherical colloid particles, and to compare these calculated values with those obtained by Ghosh and Rakshit by electro-osmotic measurements.

The total potential energy V is calculated from the following expression:

$$V = V_R + V_A \qquad (1)$$

where V_R and V_A are the repulsive and attractive potential energies of interaction of two colloid particles respectively.

Derjaguin[2] has shown that for small surface potential ψ_0 and large values of κa the repulsive energy V_R of interaction between two colloid particles, each of radius a, can be calculated from the following expression:

$$V_R = \frac{Da\psi_0^2}{2} \ln \left[1 + \exp\{-\tau(S-2)\}\right] \qquad (2)$$

where D is the dieletric constant and τ and S are equal to κa and R/a respectively, where R is the distance between the centres of two colloid particles.

Hamaker's[3] expression for the attractive energy V_A of interection of two spherical particles can be written in the following form:

$$V_A = -\frac{A}{6}\left[\frac{2}{S^2-4} + \frac{2}{S^2} + \ln\frac{S^2-4}{S^2}\right] \qquad (3)$$

where A is the Van der Waals–London constant.

In order to calculate the values of V from equations 1, 2 and 3 it is necessary to know the values of κ, a and A. The values of κ have been calculated from the following expression:

$$\kappa = \sqrt{\frac{4\pi\varepsilon^2 \Sigma n_i z_i^2}{DkT}} \qquad (4)$$

where ε is the electronic charge, D the dielectric constant of the medium, k the Boltzmann constant, n_i the number of ions of the ith type per c.c. of the solution and z_i is its valency. Taking $T = 35°$ C. and $D = 75$, the values of κ at flocculating concentrations of electrolytes are calculated from equation 4 for a titanic oxide sol[1] and given in column 3 of Table 1.

Table 1

Titanic oxide sol

Electrolyte	Concentration of electrolyte ($N \times 10^4$)	$\kappa \times 10^{-6}$	ζ observed (mV.)	ζ calculated (mV.)
$K_4[Fe(CN)_6]$	2·86	0·8847	10·3	13·94
Na_3-citrate	2·72	0·7716	9·0	12·93
Na_2SO_4	4·5	0·8596	12·3	13·72
NaCl	103·00	3·357	28·2	28·2
$K_4[Fe(CN)_6]$ + NaCl	1·43 51·50	2·456	25·0	23·97
Na_3-citrate + NaCl	1·33 51·5	2·435	24·8	23·85
Na_2SO_4 + NaCl	2·237 51·5	2·45	24·2	23·95
Na_2SO_4 + NaCl	4·114 5·15	1·114	16·1	15·86

Since the colloid particles increase in size rapidly before they finally flocculate, the value of a at the point of flocculation is very high and has been arbitrarily taken to be equal to 1×10^{-4} cm. for this calculation. It can also be shown that the calculation of the ζ-potential is not appreciably affected by even a large variation of a from this value.

The value of A has been evaluated from an analysis of the potential energy curves. Identifying ψ_0 with ζ and taking $\zeta = 28{\cdot}2$ mV., $\kappa = 3{\cdot}357 \times 10^6$ and $a = 1 \times 10^{-4}$ cm. for sodium chloride, the values of V

for different S values are calculated from equations 1, 2 and 3 for a particular value of A. A curve is drawn by plotting the values of V against S. Different curves are obtained for different values of A. According to Verwey and Overbeek[4] the transition between stable and flocculated systems is represented by the potential curve with a weak potential barrier for which the top coincides with the horizontal axis. Therefore the value of A for which the maximum of the potential energy curve just touches the S-axis represents the value of A for this particular sol. The value of A evaluated from the graph is found to be $3{\cdot}884 \times 10^{-13}$. In the evaluation of A by the above method it has been assumed that the measured ζ-potential of the coagulum in presence of 0·0103 N sodium chloride represents the true ζ-potential of the colloid particles.

After determining the value of A by the above method the ζ-potential of the flocculated particles for any other electrolyte is determined by drawing similar potential energy curves. Knowing the value of $\varkappa$ from equation 4 and taking $A = 3{\cdot}884 \times 10^{-13}$ and $a = 1 \times 10^{-4}$ cm. a total potential energy curve is drawn for a particular value of ζ. Different curves are obtained for different values of ζ. The value of ζ for which the maximum of the total potential energy curve just touches the S-axis represents the ζ value for this particular electrolyte. The ζ values evaluated by this process for different electrolytes are given in column 5 of Table 1.

The ζ values evaluated from the potential energy curves are found to be very close to the ζ values obtained by electro-osmotic measurements. The above agreement between the experimental and calculated ζ values give direct support to Verwey and Overbeek's theory.

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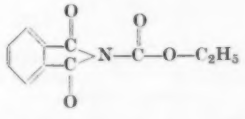
Synthesis of Phthaloyl Amino-Acids under Mild Conditions

The synthesis of phthaloyl amino-acids, using amino-acids as starting material, often proceeds under rather drastic conditions[1]. Phthaloyl amino-acid esters, however, can be prepared under less drastic conditions[2–4].



We found that N-carboethoxy phthalimide is an excellent reagent for the preparation of phthaloyl amino-acids under mild conditions. The reaction, which is carried out in aqueous solution at room temperature, applies to many functional groups.

The reagent is easily prepared from potassium phthalimide and ethyl chlorocarbonate in dimethyl formamide or from phthalimide, triethylamine and ethyl chlorocarbonate in the same solvent. It is easily crystallized from ethanol or isopropyl alcohol.

To 30 ml. of water (temp. 17°–20°) 1·5 gm. of glycine (0·02 mole), 5·75 gm. sodium carbonate 10 aq. (0·02 mole) and 4·5 gm. of N-carboethoxy phthalimide were added. The mixture was stirred for about 15 min. The solution was then filtered and the filtrate acidified. After crystallization from water and drying, 3·72 gm. of phthaloyl glycine (yield: 90·5 per cent) m.p. 191° were obtained.

With only small modifications of the above-mentioned procedure, the compounds presented in Table 1 were prepared.

Table 1

Compound	Yield	m.p.
Phthaloyl-β-alanine	91 per cent	152°
Phthaloyl-L-glutamic acid	65 per cent	160°
Phthaloyl-DL-serine	95 per cent	152°
Phthaloyl-L-asparagine	85 per cent	199°
Phthaloyl-DL-phenylalanine	90 per cent	178°
ε-phthaloyl-L-lysine*	85 per cent	232°
Phthaloyl-DL-methionine	96 per cent	102°
Diphthaloyl-L-cystine	92 per cent	120°
Phthaloyl-L-leucine	92 per cent	110°

* Prepared from the copper complex of lysine.

Phthaloyl-L-glutamic acid prepared by this method has the same rotation as reported by Clark-Lewis and Fruton[5]. After removing the phthaloyl groups in diphthaloyl-L-cystine, the cystine thus obtained (without further purification) in a yield of 85 per cent has the same rotation as our starting material, 208° in 1 N hydrochloric acid.

From these facts we conclude that the introduction of the phthaloyl group by our method respects the optical activity of the amino-acids.

Details and proposed mechanism will be published elsewhere (*Rec. Trav. Chim.*).

I would like to thank G. I. Tesser for the benefit of many discussions.

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Paper Chromatography of Acidic Carbohydrates

Cationic detergents, such as cetyltrimethylammonium bromide ('Cetavlon') and cetylpyridinium chloride, have been used extensively in recent years for the fractionation of polysaccharide mixtures[1–4]. The quaternary ammonium cations present in these detergents form salts with acidic polysaccharides which often have solubility properties quite different from those of the usual metallic salts; for example[1], cetyltrimethylammonium chondroitin sulphate is soluble in aliphatic alcohols up to n-pentanol and is insoluble in water. Many purification procedures have utilized the water-insolubility of these quaternary ammonium polysaccharide salts, but surprisingly, no serious attempt seems to have been made to exploit their solubility in organic solvents. This communication describes some preliminary experiments directed towards developing convenient methods for the separation of acidic carbohydrates, making use of the solubility of their cetylpyridinium salts in organic

solvents. Current interest in these laboratories in the purification and identification of sugar sulphates, suggested that it would be useful if this property could be utilized in a method for the paper chromatography of these substances. It has been found that when a small amount of detergent is incorporated into the mobile (organic) phase of certain solvent systems for paper chromatography, the R_F values for sulphated derivatives are increased by a factor of about ten, and some useful separations can be achieved. Two such systems are :

(*a*) *Butanol/ethanol/water* (3 : 1 : 1 *by vol.*; 100 *ml.*) *plus cetylpyridinium chloride* (3 *gm.*). The R_F values for some sugar sulphates in this solvent system are given in Table 1. The mixture obtained by the direct sulphation of glucose (with pyridine–sulphur trioxide) was resolved into at least five components (Table 1). The major monosulphated component had the same R_F value as glucose-6-sulphate, thus confirming previous results[5]. Small quantities of two other glucose monosulphates were detected, one of which had the same R_F value as glucose-3-sulphate.

Table 1. R_F Values of Some Sugar Sulphates*

	R_F values*	
	Solvent (*a*)†	Solvent (*b*)†
Glucose	0·21	0·18
Glucose-3-sulphate	0·67	0·85
Glucose-6-sulphate	0·41	0·77
Glucose disulphates	1·00‡	1·00‡
Products from direct sulphatation of glucose	0·21, 0·41, 0·55, 0·67, 1·00	0·18, 0·77, 0·85, 1·00

* R_F values are measured relative to the rate of movement of the slower of the two solvent fronts.

† Development period for solvent (*a*) 16 hr.; solvent (*b*) 3 hr.

‡ Travels as an extremely compact zone.

(*b*) *Water-saturated methyl ethyl ketone* (100 *ml.*) *plus cetyl pyridinium chloride* (3 *gm.*). Although this solvent system is less useful than system (*a*) for the separation of isomeric glucose monosulphates, it does possess certain advantages. Thus, simple mixtures can be resolved very rapidly; for example, glucose and glucose monosulphate are well separated by a 15-min. development using the ascending technique. (Equilibration of the paper against the solvent vapour prior to development may, however, be advisable when more complex mixtures are to be analysed.) The detection of acidic sugar derivatives in a mixture (for example, a polysaccharide partial hydrolysate) should be rapid and simple since only these substances will move faster than the parent monosaccharide when the detergent is incorporated into the solvent, and slower when it is omitted.

For the location of sugar zones after irrigation of the chromatogram, two methods have been found useful. The silver nitrate–sodium hydroxide reagents[6] can be used if the paper is first rinsed with chloroform to remove the detergent, or *p*-anisidine hydrochloride[7] can be applied directly to the dried chromatogram and, after heating, the spots observed in ultra-violet light.

This work is being continued with the view of developing analogous methods for the paper chromatography of other acidic carbohydrates.

I am grateful to Mr. T. P. Williams and Mr. K. O. Lloyd for the samples of sugar sulphates used in this work, to the Department of Scientific and Industrial Research for financial assistance, and to Prof. Stanley Peat and Dr. J. R. Turvey for their interest and encouragement.

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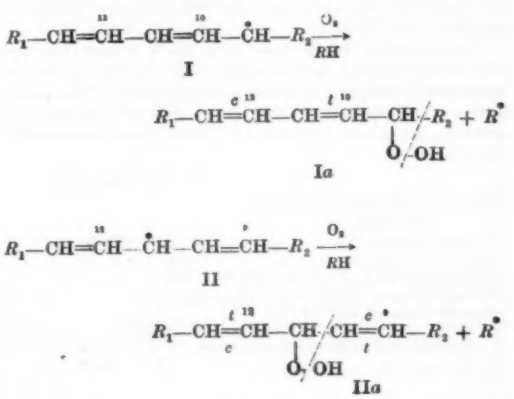
The Stereo-configuration of 2,4-Decadienals isolated from Oils containing Linoleic Acid

According to Farmer's theory (see Bolland[1] and Bateman[2]), the following isomeric hydroperoxides (I*a*, II*a*, and III*a*) are formed in the autoxidation of linoleic acid, from the mesomeric linoleic acid free-radicals (I, II and III) :



$R_1 = CH_3—(CH_2)_4—$ RH = linoleic acid molecule
$R_2 = —(CH_2)_7—COOH$ $R^\bullet$ = linoleic acid radical
t = *trans*, *c* = *cis*.

The theoretical stereoconfiguration of the possible isomers has been indicated by Nichols *et al.*[3].

Swift *et al.*[4] have isolated from cottonseed oil (which contains approx. 45 per cent linoleic acid) in various stages of autoxidation 2,4-decadienal, 2-octenal, and hexanal. Recently, Patton *et al.*[5] have published results on the occurrence of 2,4-decadienal in several oils and fats. The formation of these decomposition products can be readily explained by means of the above-mentioned theory. From the

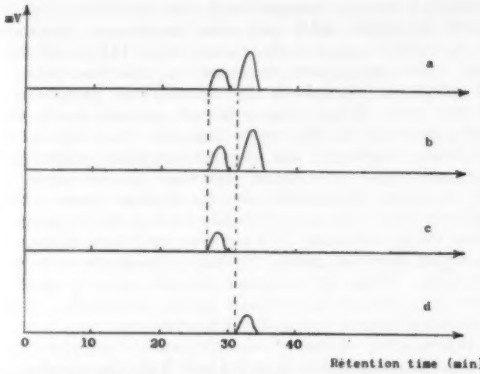


Fig. 1. Gas-liquid chromatography of: *a*, palm oil decomposition product (detail); *b*, groundnut oil, ditto; *c*, synthetic 2(*trans*)-4(*cis*)-decadienal; *d*, synthetic 2(*trans*)-4(*trans*)-decadienal. Carrier, 'Celite 545'; immobile phase, silicone oil (30 per cent); mobile phase, nitrogen; pressure, 50 cm. mercury; gas velocity, 50 ml./min.; temperature, 137° C.; length of column, 90 cm.; detector, gas density balance

relevant literature, however, no information can be obtained on the stereo-configuration of the unsaturated aldehydes obtained, notably 2,4-decadienal.

From our investigations with palm oil and groundnut oil it appeared that on subjecting the volatile decomposition products to gas-liquid chromatography[6], two aldehydes could be isolated, which were identified as two isomeric 2,4-decadienals (Figs. 1*a*, *b*; 2*a*, *b*). By partition chromatography of the dinitrophenylhydrazones, following the method of Miss Kramer and van Duin[7], the 2,4-decadienal structure could be established, but not the stereo-configuration.

Two isomeric 2,4-decadienals of known stereo-configuration were synthesized. Using Brown and Subba Rao's method[8] 2(*trans*)-4(*cis*)-decadienal was prepared from 2(*trans*)-4(*cis*)-decadienoic acid. This acid was isolated, in a yield of 3–5 per cent, from *Stillingia* oil, the oil from the seeds of *Sapium sebiferum* Roxb. of which the stereo-configuration has been established by Crombie[9]. By application of Doebner's method (cf. Forss and Hancox[10]) 2(*trans*)-4(*trans*)-decadienal was synthesized, starting from hexanal. As the synthetic products and the two aldehydes isolated from volatile decomposition products gave similar results in the gas-liquid chromatographic analysis (Figs. 1*c*, *d*) and in the infra-red analysis (Figs. 2*c*, *d*), the conclusion is justified that the two natural 2,4-decadienals obtained possess the 2(*trans*)-4(*cis*) and 2(*trans*)-4(*trans*) stereo-configuration. From the gas chromatogram these *trans,cis* and *trans,trans* compounds were found to be present in the ratio of 28 : 72.

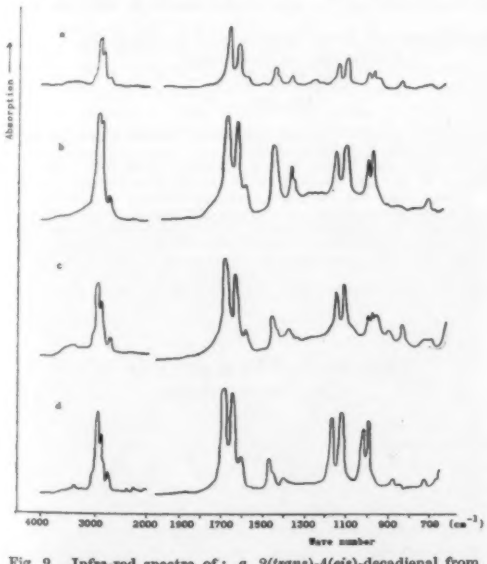


Fig. 2. Infra-red spectra of: *a*, 2(*trans*)-4(*cis*)-decadienal from palm oil (first peak in Fig. 1*a*); *b*, 2(*trans*)-4(*trans*)-decadienal from palm oil (second peak in Fig. 1*a*); *c*, synthetic 2(*trans*)-4(*cis*)-decadienal (cf. Fig. 1*c*); *d*, synthetic 2(*trans*)-4(*trans*)-decadienal (cf. Fig. 1*d*).
To record these spectra, a 'Unicam' 100, double-beam infra-red spectrophotometer was employed (rock-salt prism, microplate technique)

The above affords new, indirect proof that the (I*a*) isomer of linoleic acid hydroperoxide is formed as one of the primary autoxidation products as found spectrophotometrically by Sephton and Sutton[11]. Then it must be assumed that isomer (I*a*) serves as precursor for 2(*trans*)-4(*cis*)-decadienal. The more stable 2(*trans*)-4(*trans*)-decadienal can be formed, either as a secondary reaction product during further rearrangement of isomer (I*a*) to 9-hydroperoxy-10(*trans*)-12(*trans*)-decadienoic acid, or as a result of the instability of 2(*trans*)-4(*cis*)-decadienal. More detailed results will be published elsewhere.

We wish to thank Mr. J. H. Recourt for his assistance in carrying out the gas-liquid chromatography experiments. Further, our thanks are due to Unilever Research Department, Port Sunlight, for supplying the sample of *Stillingia* fatty acids and to the management of Unilever N.V., Rotterdam, for permission to publish these data.

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Effect of Flow-Rate and Sample-Size on Column Efficiency in Gas-Liquid Chromatography

There have been a number of investigations as to the effect of carrier gas flow-rate on separation and column efficiency in gas-liquid chromatographic systems[1–4]. Many of the results present contradictory pictures of the effect exerted by this parameter. Keulemans[1] states, "for a certain set of operational conditions there is an optimum linear gas velocity, but the optimum is very flat and the gas rate is consequently not a highly critical parameter". James and Martin[2] have shown that column efficiency is sensitive to flow-rate and that a decrease in flow-rate increases column efficiency. Their results show a

27 per cent increase in column efficiency when the flow-rate was decreased from 24 to 10 ml./min. Bethea and Wheelock[3] reported that column efficiency, as measured by the H.E.T.P., is approximately tripled by raising the flow-rate from 5 to 27·5 ml./min. They found that increasing the flow-rate beyond the latter point gave only meagre increases in efficiency.

Mellor[4] found the critical flow-rate to be independent of temperature, pressure, column length, and load size, but dependent on column diameter and particle size of the column packing support.

During an investigation of the separation of two terpene hydrocarbons, α-terpinene and D-limonene, by means of gas-liquid chromatography, it was of interest to determine whether better separation could be achieved by selection of an optimum carrier gas flow-rate and sample size. It is a well-established fact that the efficiency of separation in a gas-liquid chromatographic column improves as the size of the sample is reduced[1,2]. Thus, with low column-load and optimum gas-flow, a high efficiency should be achieved.

The apparatus employed to separate α-terpinene from D-limonene was an 'Aerograph' model *A*-90-*C* (manufactured by Wilkins Instrument and Research, Inc., Walnut Creek, California). The column was constructed of stainless steel tubing, ¼ in. outside diameter and 10 ft. in length. The support material used was 'Sil-O-Cel' *C*-22 diatomaceous earth firebrick (30–60 mesh). Fractions of this were sieved to size and further graded by sedimentation in water, after which they were dried at 150° for 16 hr. The liquid phase was introduced by deposition from benzene solution, the packing then being heated in a stream of helium at 150° for 16 hr.

The column was contained in an air thermostat; the carrier gas flow (helium) was controlled by a precision reducing valve and measured by a soap bubble flow meter situated at the column outlet. Temperature and flow control were sufficiently good to make possible the use of a four-channel, hot-wire katharometer that proved virtually noiseless when connected with a 1 mV. recording potentiometer. Sample injection was made with a microsyringe, and the sample preheater was maintained at 150°.

Separations were carried out with the aid of a stationary liquid phase consisting of *LAC*-4-*R*777 (the succinate polyester of diethylene glycol)[5]. This material was applied to the solid support in the amount of 25 per cent w/w. Column temperature was maintained at 100°, and recorder chart speed was 30 in./hr. The column outlet was maintained at atmospheric pressure.

Flow-rates varied from 20 to 145 ml./min. of helium. Table 1 illustrates the effect of flow-rate and sample-size on column efficiency under the experimental conditions described above. The number of theoretical plates was calculated by means of the equation[5]:

$$\text{Number of theoretical plates} = 16\left[\frac{\text{Retention volume}}{\text{Peak width}}\right]^2$$

As the number of theoretical plates can vary with the nature of the component[5], a calculation was made for both peaks on each chromatogram, and the average of the two values is presented in Table 1. It should be noted that the theoretical plate values for both peaks varied in a manner similar to that displayed by the average of the two values.

With a 0·1-μl. sample load, the theoretical plate value increased 63·8 per cent when the flow-rate of the carrier gas was decreased from 145 to 45 ml./min. An additional decrease in the flow-rate to 20 ml./min. decreased the theoretical plate value 43 per cent. Employing a 0·3-μl. sample load, this same decrease in flow-rate, that is, from 145 to 45 ml./min. increased the theoretical plate value 46·9 per cent. and a further decrease in flow-rate to 20 ml./min. decreased the theoretical plate value 26·3 per cent. At a sample load of 0·5 μl. the theoretical plate value increased 25·8 per cent and then decreased 24·6 per cent in going through the same flow-rate changes. Thus at constant sample size, a plot of flow-rate versus theoretical plate value for these systems shows a pronounced maximum.

When one operates at optimum flow-rate and varies the sample size from 0·1 to 0·3 μl., the theoretical plate value decreases by 12·5 per cent. Increasing the sample size to 0·5 μl. reduces the theoretical plate value by 39·5 per cent.

Bethea and Wheelock[3] have noted that as the flow-rate is increased the optimum sample size for maximum column efficiency increases. It may be noted that the optimum flow-rate appears to shift toward 60 ml./min. at the higher column load (Table 1).

Table 1

Flow-rate (ml./min)	Theoretical plates		
Sample size	0·1 μl.	0·3 μl.	0·5 μl.
20	1499	1508	1096
30	1967	1751	1167
45	2144	1905	1282
60	2049	1851	1366
74	—	—	1352
92	1592	1601	1188
145	1309	1297	1086

Investigations of the effects of these operational parameters are being continued and will be published in the future.

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Cathode-Ray Presentation of Chromatograms

From the results of investigations on nylon capillary columns[1], it became apparent that there were two possible applications for the capillary system. The columns could be used to produce very high efficiencies with elution times of 10–60 min., or the high efficiencies could be sacrificed and chromatograms developed in a matter of seconds, with moderate efficiencies of perhaps 5,000 or 10,000 plates. Under these conditions it was found that a chromatogram of a mixture boiling over a range of about 100 deg. C. would be developed in 60 sec., the early peaks being eluted during a period of about

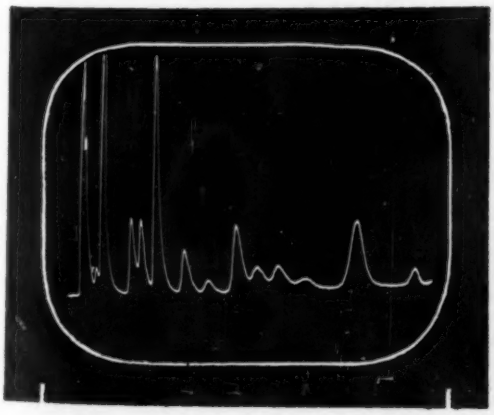
0·5 sec., but it was necessary to use a high-speed recording system with time constants of the order of milliseconds. Such a performance cannot be achieved by the normal potentiometric recorder, and the oscilloscope is an obvious alternative method of presenting the chromatogram.

In addition to the improved speed of response, an oscilloscope is also cheaper to manufacture. The complete chromatographic equipment, incorporating the column, detector and ancillary equipment, could be manufactured for less than the price of a single recorder.

With such an apparatus, it would be possible to analyse a volatile mixture in a matter of seconds, and it could be used as a laboratory instrument for routine analysis. By use of an automatic injection system synchronized with the scan, it would be possible to monitor the product from a plant every minute. Another interesting application would be its use in the investigation of reaction kinetics, where the complete analysis of a volatile mixture in a matter of seconds would be a distinct advantage.

The construction of a suitable display system is comparatively easy. The results shown below were obtained from a normal television amplifier system and power supply, coupled to a 14-in. magnetically deflected cathode-ray tube coated with a phosphor having a 200-sec. persistence. The micro-argon detector devised by Lovelock was used, in conjunction with a modified impedance converter of extremely low time-response. The scan was obtained by a linear-wound potentiometer driven by a constant-speed motor. The column used was 70 ft. of 0·020 in. diameter nylon tubing coated with a total of 8 mgm. of 'Apiezon Oil *A*' as liquid phase, and was run at an inlet pressure of 9 lb./sq. in. A 100 : 1 diluting system was used on the injection, in order that a sufficiently narrow band of solute could be placed on the column[2]. The total charge on the column was about 2 μgm. and the sensitivity of the detector was of the order of 10^{-11} gm./c.c.

A photograph of the trace obtained from the analysis of a petroleum ether mixture is shown in Fig. 1. The first two peaks, *iso*-butane and *n*-butane, are just resolved, the third high peak is *n*-hexane and the last peak is methyl*cyclo*hexane. The total scan of the tube was 50 sec. The first peak, *iso*-butane, was eluted in 31 sec. measured from the time of



50 sec.

Fig. 1. Chromatogram from cathode-ray tube

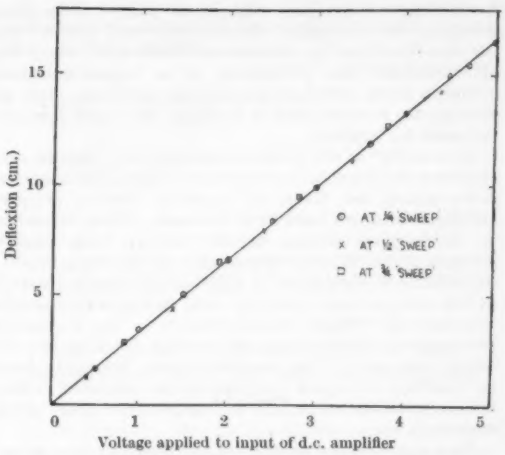


Fig. 2. '*Y* deflexion'/voltage input to d.c. amplifier

injection, and the elution curve itself was developed over a period of 0·5 sec. *n*-Hexane was eluted in 40 sec. and *n*-heptane in 80 sec. The efficiency for *n*-hexane on this chromatogram is between 5,000 and 6,000 plates. This is not obvious because the injection is not shown, as it occurred about two-thirds of the way through the previous scan.

The amplifying system was adjusted to give a linear response between voltage input on the d.c. amplifier and centimetres deflexion on the *Y*-axis of the oscilloscope. Thus, providing the micro-argon detector is adjusted to give a linear response, then quantitative results can be obtained from the chromatogram. The relationship between centimetres deflexion and voltage input to the d.c. amplifier is shown in Fig. 2.

The instrument is still under development, and results have shown that higher efficiencies and even shorter times of elution are practical. However, these early results clearly indicate that the oscilloscope system is an alternative method for presenting chromatograms. Full details of this work will be published elsewhere.

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BIOCHEMISTRY

Metal-Organic Complexes formed during the Treatment of Wood with Metal Salts

A RECENT communication by Belford, Cook and Nevard[1] dealt with the subject of metal-cellulose complexes formed during the treatment of Douglas fir wood with various preservative processes comprising solutions of inorganic salts or mixtures of these. The authors claim that these are formed at

room temperatures, and that, in the case of the copper complex, the process by which it is formed differs from the one described by Abrams and Bottoms[2], who have also claimed the formation of a copper-cellulose complex when cellulose-containing materials, such as cotton, are treated with a solution of copper formate followed by heating.

In a study[3] of the process described by Abrams and Bottoms we have been unable to confirm the observations which led them to conclude that a copper-cellulose complex had been formed. Thus, bleached or unbleached cotton fabric treated with copper formate solution according to this method was readily dispersible in a solution of cuprammonium hydroxide of the composition normally used in dispersing textile cellulose for fluidity measurements[4]; the copper in the treated fabric could be readily leached out by dilute ammonia; the decomposition, brought about by heating, of copper formate on the bleached cotton fabric, gave the same colour change when glass fabric was used.

In a study of the kinetics of the thermal decomposition of metallic formates, Kornienko[5] found that the main products of the reaction were a metal oxide and products of the decomposition of formic acid, and that where free metal was present in the reaction products it could be attributed to the reduction of the oxides by the gaseous decomposition products, notably carbon monoxide and hydrogen. It is of interest to note that the mechanism postulated by this author would make it possible for cuprous oxide to be one of the reaction products. While the evidence from X-ray diffraction data for the presence of cuprous oxide in our fabric treated with cupric formate followed by heating is inconclusive, it does not preclude its presence.

Moreover, we found that the resistance to microbiological attack shown by cotton fabric treated with cuprous oxide was similar to that shown by the copper formate treated fabric containing a similar percentage of copper. Thus fabrics containing cupric formate and cuprous oxide at concentrations of 0·43 per cent and 0·26 per cent respectively gave essentially the same resistance in the soil burial test, namely, 6–10 per cent loss of strength for the former and 13 per cent loss for the latter.

In an earlier communication[6] Cook and Nevard refer to the copper formate process for the preservation of wood as described by Walton-Smith *et al.*[7], this being the same process described by Abrams and Bottoms[2]. Here, it is claimed by Walton-Smith *et al.* that a copper-cellulose complex is formed in the wood, and it is stated by Cook and Nevard in the article referred to above that their results prove conclusively the existence of this copper-cellulose complex. However, in the later communication[1], these latter authors give data which, they claim, indicate that the copper-cellulose complex which they believe to have been formed in their experiments is not of the same type as the one postulated by Abrams and Bottoms.

If by 'copper-cellulose complex' Cook and Nevard mean an association of copper and substantially pure cellulose we would take exception to this view, and we would suggest that the complexes formed be regarded as predominantly the product of the interaction of the metal ions with acidic, cation-binding, non-cellulose constituents of the wood. Indeed, in view of the close association of these non-cellulosic materials with the cellulose of wood, it is difficult to envisage the formation of a pure metal-cellulose complex in the amounts claimed by Cook and Nevard under the conditions of their experiments.

It is well known that 'native celluloses' show marked, but varying, tendency to take up metal cations after treatment with acid followed by washing. This effect has been examined by a number of workers and it has been shown that although a highly purified cellulose consisting substantially of α-cellulose has a small cation-binding power, relating probably to the presence of acidic end groups[8], this capacity to bind cations is greatly increased as we approach more nearly to the 'native celluloses' which are known to contain non-cellulosic constituents such as pectins, uronic acids and lignin[9].

In some of their treatments of wood with inorganic salts, Cook and Nevard observed heavy incrustations of electron-opaque material. They also observed that certain fragments of treated wood, whether showing incrustations or not, gave sharp electron diffraction diagrams, characteristic of the treatment; that in the case of copper treatments, these diagrams did not resemble any of the X-ray patterns for the copper salts employed and also that the diffraction diagram faded fairly rapidly with increasing intensity of the electron beam. All these observations are consistent with our view that the complexes formed consist of metal cations bound on the acidic groups of non-cellulosic constituents of the wood.

Such a view is also consistent with the observation of Belford, Cook and Nevard that the reaction leading to the formation of the complexes takes place at room temperature.

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Mediation of the Genetic Control of Protein Synthesis in Cell-free Extracts of *Neurospora crassa*

I have recently demonstrated the development of tryptophan synthetase activity by cell-free extracts from conidia of *wild-type Neurospora crassa*. The process appeared to be one of synthesis of enzyme protein, and was associated with the net increase in the protein content of the system[1]. Similar extracts prepared from the *td*$_2$ *tryptophan-less* mutant strain neither contained nor developed any detectable tryptophan synthetase activity, but did catalyse a net synthesis of protein.

The defective component of the mutant extract has been located in the 'soluble' fraction not sedimented by centrifuging at 105,000 *g* for 1 hr. As shown in Table 1, when a mutant 'particle' fraction was supplemented with *wild-type* 'soluble' fraction, considerable tryptophan synthetase activity was developed. Con-

Table 1. Location of the Genetic Defect in Extracts of *td*$_2$ Mutant Strains

Tryptophan synthetase activities (units/ml. of conidial ext.)

'Soluble' Fraction 'Particle' Fraction	Wild Type Wild Type		Mutant Mutant		Wild Type Mutant		Mutant Wild Type	
Exp. No.	Initial	Final	Initial	Final	Initial	Final	Initial	Final
1	0·21	0·37	0	0	0·21	0·30	0	0
2	0·52	0·72	0	0	0·56	0·79	0	0
3	0·20	0·37	—	0	—	0·30	—	0
4	0·30	0·47	0	0	—	0·44	0	0
5	0·17	0·34	0	0	0·19	0·30	0	0

versely, a mixture of *wild-type* 'particles' with mutant 'soluble' fraction failed to develop any detectable activity.

My method of preparing conidial extracts[1] (see below) could cause contamination by material from the cell nuclei. However, in view of the short duration of the experiments, it seems unlikely that this material contributes significantly to the development of tryptophan synthetase activity observed. Further, extracts prepared from *wild-type* mycelium by milder homogenization procedures also readily form active enzyme (unpublished results). I therefore believe that the defective component of the *td*$_2$ mutant extracts is some constituent of the cell 'sap'. Certainly, it would seem that the primary genetically determined defect in the mutant conidium is not located in the 'particulate' fraction corresponding to the microsomes of mammalian cells.

This conclusion has important implications for our understanding of the manner in which complex gene loci control the formation of specific enzymes, and of the mechanism of action of suppressor genes.

Extracts were prepared from conidia harvested from the growth on solid synthetic medium[1]. *Wild-type* cultures were grown at 30° C. for 4 days, and incubated at room temperature (about 20° C.) for a further 4 days. Mutant cultures were grown at 30° C. for 7 days on medium supplemented with 0·04 per cent (w/v) DL-tryptophan, and incubated a further 7 days at room temperature. The washed and chilled conidia were ground with powdered glass, extracted with cold *M*/20 sodium potassium phosphate buffer (*p*H 6·5) containing 20 per cent (w/v) sucrose, and separated into 'soluble' and 'small particle' fractions as described in detail elsewhere[1].

The reaction system contained 'soluble' fraction, 'small particle' fraction, 4 per cent (w/v) Difco 'Bacto Casamino Acids', 6·4 mgm. L-cysteine hydrochloride, 50 mgm. hexose diphosphate magnesium salt, 4 mgm. diphosphopyridine nucleotide, 0·4 mgm. calcium chloride, $1{\cdot}26 \times 10^{-1}$ *M* magnesium sulphate and 42 mgm. of polyvinyl acetate (as a finely dispersed emulsion) in a final volume of 28 ml. In experiment 5 a mixture of 8 mgm. of L-proline plus 3·2 mgm. of each of L-threonine and L-valine replaced half the casein hydrolysate.

Tryptophan synthetase enzyme was precipitated from samples of the reaction system, taken before and after incubation for 1 hr. at 30° C., by a modification[1] of the procedure of Suskind and Kurek[2], and enzymic activity was assayed by a modification[1] of Yanofsky's method[3]. Total protein nitrogen was determined by standard micro-Kjeldahl techniques after precipitation with trichloracetic acid, and extraction of nucleic acids and lipids[1].

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Anærobic Glycolysis of Dispersed Cell Suspensions from Normal and Malignant Tissues

In studies of cellular metabolism, the potential advantages of using dispersed cell suspensions rather than tissue homogenates or slices have led a number of workers to investigate the methods of preparation and properties of cell suspensions from various tissues[1–5]. Liver parenchymal and kidney epithelial cell suspensions prepared by mechanical disruption of the tissue, with or without prior perfusion of the organ, appear to be morphologically intact when examined by either light or electron microscopy[1,2]. These cell suspensions, however, are apparently metabolically different from tissue slices, since they show negligible endogenous respiration which cannot be restored by glucose or a number of protein and non-protein factors[2,3], and have lost the ability to convert intracellular carbohydrate to lactic acid[3]. The oxidation of succinate by these cells, however, has been reported to proceed at a rate exceeding that observed with slices[2,3], and is independent of exogenous cytochrome *c* (ref. 4). Liver cell suspensions have also been found to incorporate glycine at a rate seven-fold greater than that observed with homogenates[6].

We have found that cell suspensions from a number of solid and ascitic animal tumours metabolize glucose either aerobically or anærobically, whereas isolated liver and kidney cells do not. This difference in behaviour between these normal and tumour cell suspensions was found to be due to a nearly complete loss of some of the enzymes of the Embden–Meyerhof system from the kidney and liver cells, but not from the tumours tested.

Rat liver and kidney cells were prepared by Anderson's procedure[1], namely, perfusion of the organ with calcium-free Robinson's solution containing 0·027 *M* citrate, followed by dispersion of the tissue in a loose-fitting homogenizer. The cells were then washed twice with calcium-free Robinson's solution. Cells from solid tumours were prepared in the same fashion, except that the tissues were dispersed without prior perfusion; ascites cells were diluted with the perfusion medium and washed as above. The yield of intact cells varied with source of tissue, but was in close agreement with published results[1]. Suspensions of dispersed cells from the solid Novikoff hepatoma and both the Novikoff and Ehrlich ascites tumours were found to grow at a rate equal to, or better than, untreated tumour mince, or whole ascites fluid, respectively, when transplanted, indicating no loss of viability during isolation.

The lack of respiration of liver and kidney cells in an unfortified system with or without glucose was con-

Table 1. Anærobic Glycolysis of Cell Suspensions

The reaction system (3·0 ml.) contained: 7·5μ moles phosphate (*p*H 7·4), 20 μmoles magnesium chloride, 49·5 μmoles potassium bicarbonate, and 150 μmoles potassium chloride; 1·0 μmole adenosine diphosphate (ADP), 1·0 μmole diphosphopyridine nucleotide (DPN), and 50 μmoles glucose where indicated. The vessels were gassed with 95 per cent nitrogen–5 per cent carbon dioxide for 10 min. Incubation, 1 hr. at 30° C. The values are μmoles lactic acid/hr./mgm. nitrogen.

Tissue	Minus ADP and DPN		Plus ADP and DPN	
	Endogenous	+Glucose	Endogenous	+Glucose
Liver	0	0	0	0
Kidney	0	0	0	0
Thymus	0·2	2·0	0·9	2·2
Novikoff hepatoma (solid)	0·3	2·0	0·9	5·0
Murphy lymphosarcoma	0·3	4·4	0·3	4·6
Walker-256	0·3	1·5	0·8	4·6
Gardner lymphosarcoma	0·7	5·3	0·9	8·0
Novikoff hepatoma (ascites)	0·8	4·6	1·0	5·5
Ehrlich ascites	0·5	5·6	0·6	6·0

firmed, whereas tumour cell and thymocyte suspensions show both endogenous and glucose-stimulated respiration. Succinate was oxidized by all cell suspensions studied, and the rate could be stimulated by addition of adenosine diphosphate. Preliminary results of a study of the oxidation of a variety of substrates by these cell suspensions indicate that adenosine diphosphate and diphosphopyridine nucleotide are required for maximal activity.

The results of a comparison of anærobic glycolysis by cell suspensions from various tissues are shown in Table 1. Liver and kidney cells did not catalyse anærobic glycolysis, as measured in terms of lactic acid production/hr./mgm. nitrogen from glucose. There was, however, a small but measurable release of carbon dioxide when measured manometrically. Fortification of the medium with adenosine diphosphate and diphosphopyridine nucleotide did not restore the glycolytic ability of these cells, indicating that the metabolic alteration was not caused by leakage of these co-factors. No activity could be demonstrated using the system described by LePage[7], which was designed to demonstrate glycolysis with homogenates of normal tissues. Thymocytes and all the tumours tested catalysed an active glycolysis, both from endogenous substrates and glucose, which could be stimulated by adenosine diphosphate and diphosphopyridine nucleotide with some tumour cell suspensions.

The inability to produce lactic acid from glucose in liver and kidney cells has been demonstrated to be due to the loss of at least two key enzymes of the glycolytic pathway. In Table 2 the specific activity of aldolase and lactic dehydrogenase of the original dispersed tissue, prior to centrifugation, and the final washed-cell suspension are compared, since measurements of total activity in the fractions are not significant due to variations in the yield of cells from the various tissues. Isolated liver and kidney cells contained negligible aldolase and lactic dehydrogenase, whereas cells from thymus and the tumours retained a significant quantity of both enzymes. The enzymes lost from the normal cells were quantitatively recovered in the wash fluids. Glycerophosphate dehydrogenase has also been found to be completely leached out of the liver and kidney cells. This leakage of enzymes has been demonstrated with liver and kidney cells prepared by other techniques for dispersal[4,5]. Perfusion with various media (for example, serum albumin in saline, whole serum, tissue culture medium 199 and mouse ascitic fluid all containing citrate) did not yield cells which retained these enzymatic activities. Cell suspensions from all solid tumours with the exception of the Murphy lymphosarcoma, which requires drastic treatment for dispersal, retained the greater part of their initial content of these enzymes, as shown by a comparison of the specific activity of the dispersed tissue fraction and the washed cell suspension.

Table 2. Aldolase and Lactic Dehydrogenase Activity of Tissue Homogenates and Cell Suspensions

Aldolase activity was determined essentially by the method of Dounce *et al.* (ref. 8). Specific activity equals change in optical density (540 mμ) per mgm. nitrogen of chromogen formed in 15 min. Lactic dehydrogenase determined spectrophotometrically; specific activity is μmoles reduced diphosphopyridine nucleotide oxidized/min./mgm. nitrogen, measured as the initial rate. Values are the average of three experiments.

Tissue	Aldolase activity		Lactic dehydrogenase	
	Dispersed tissue*	Cells	Dispersed tissue*	Cells
Liver	3·8	0·1	8·6	0
Kidney	3·2	0·1	2·0	0
Thymus	2·2	1·4	3·1	1·7
Novikoff hepatoma (solid)	3·5	2·7	3·5	2·5
Murphy lymphosarcoma	11·7	4·1	3·1	1·8
Walker-256	8·8	4·8	6·0	3·2
Gardner lymphosarcoma	5·2	3·6	3·4	3·6
Novikoff hepatoma (ascites)	6·3†	13·6	4·9†	12·1
Ehrlich ascites	4·2†	4·9	5·4†	8·7

* Tissue after dispersion but prior to centrifugation.
† Total ascites fluid and cells.

Although appearing morphologically intact, liver and kidney cell suspensions have been demonstrated not to be biochemically 'intact' due to the complete loss of some soluble enzymes. The use of such cell suspensions for certain metabolic studies is thus limited. The differential leakage of some enzymes from cell suspensions of malignant tissues and thymus as compared with liver and kidney suggests a fundamental difference in the membrane structure of cells depending on the degree of organization of the tissue source.

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Spectrophotometric Determination of Tryptophan by the Method of Roth and Schuster: Some Sources of Error

In Roth and Schuster's method[1] for colorimetric determination of tryptophan in vegetable matter, the measurement of the yellow colour is made with a Pulfrich Stufenfotometer with filter *S* 43. The method has often been used, but I have been unable to find any published account of the measurements made spectrophotometrically. I have therefore tested the method for some cereals, etc., by means of a Beckman spectrophotometer, model *D.U.*

It appeared that the absorption curves for the yellow compounds produced by D,L-tryptophan and the vegetable matter, respectively, have the same form only within a limited range of wave-lengths. In preliminary attempts to account for this, it was observed that nitric oxides developed during the process of nitration (especially in the case of substances rich in carbohydrates) are dissolved in the reaction mixture, to which they impart a yellow

colour. Furthermore, it was found that lignin, which occurs in lignified vegetable matter, is nitrated, forming vividly yellow compounds, and the determination is thus distorted.

The effect of the nitric oxides was shown by the difficulty of obtaining reproducible determinations. The following experiment showed that the nitric oxides contributed to producing the yellow colour: the reaction mixture was bubbled with nitrogen dioxide at room temperature until the air above the fluid assumed a reddish-brown colour from nitric oxides. Dilutions of the solution showed that as little as 1/16 of the original quantity of nitrogen dioxide gave a measurable extinction at 450 mμ.

The occurrence of nitric oxides in the solutions for analysis can be shown by testing for nitrite (development of a red colour by diazotization of sulphanilic acid and coupling in alkaline fluid with α-naphthol), as nitrous acid develops in the solutions:

$$2\,NO_2 + H_2O \rightleftharpoons HNO_2 + HNO_3$$

The nitric oxides could be removed completely by sucking air vigorously through the filtered solutions to be analysed. The procedure only caused a slight change of the extinctions (2 per cent), for which correction can be made.

The solutions for analysis (wheat grain, wheat straw) contained nitrogen dioxide corresponding to $\frac{1}{2}$–$\frac{1}{8}$ of the nitrogen dioxide test solution.

The absorption curves for the nitrated solutions of D,L-tryptophan show a maximum at 330–340 mμ. The extinction curves for the wave-lengths of 380, 400, 420, and 450 mμ (1–10 mgm./50 ml., 1 cm. cuvettes) and 400, 410 . . . 500 mμ (5-cm. cuvettes) were all straight lines.

Samples of grain (wheat, oats, rye, barley) and proteins (gluten, albumen) on nitration yielded yellow products, the absorption curves of which for wave-lengths up to about 440 mμ were steeper than those for tryptophan, while for the range 440–500 mμ they followed those of tryptophan, indicated by the fact that the extinctions here, calculated as tryptophan, yielded the same values.

For hulls of oats and for wheat straw the results were different. On nitration they left orange-coloured precipitates, which could only with difficulty be rinsed free from colour. The solutions showed greater absorption in the range of about 470–500 mμ than tryptophan (crossing of the tryptophan curve). The extinction at 450 mμ, calculated as tryptophan, yielded unusually high values. For wheat-straw (0·5 gm. sample) a tryptophan content of 2·25 per cent was found, corresponding to 25·3 per cent tryptophan in the protein (nitrogen × 6·25).

On going through groups of substances which might be supposed to give rise to the formation of yellow-coloured nitration products, lignin attracted special attention, as this is of an aromatic character and is found in lignified vegetable matter, such as the straw and hulls of grain.

Further evidence was obtained by treatment of samples of lignin (acid-lignin). On nitration, compounds of a vividly yellow colour developed, the absorption curves of which showed the crossing of the tryptophan curve characteristic of wheat straw and oats hulls. At 450 mμ a quantity of 0·01 gm. lignin gave the same extinction (0·102) as 1 mgm. tryptophan (0·105), and Roth and Schuster's method therefore in its present form must be considered unsuitable for vegetable matter with even a slight content of lignin.

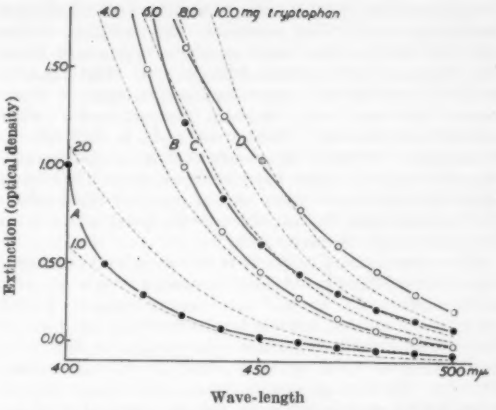

Fig. 1. Absorption curves measured with the Beckman spectrophotometer, model *D.U.*, in 5-cm. cuvettes against reaction mixture. Broken curves, D, L tryptophan. Curve *A*, nitrogen dioxide dissolved in reaction mixture; curve *B*, 1·5 gm. wheat grain; curve *C*, 1·0 gm. oats straw; curve *D*, 0·1 gm. lignin. The solutions were diluted to 50 ml. before being measured

As to other kinds of vegetable matter, etc., it is necessary to undertake the measurements at a wave-length higher than about 440 mμ, which necessitates the use of 5-cm. cuvettes. However, it will be desirable to investigate the character of compounds which cause increased absorption below about 440 mμ, as the possibility cannot be precluded that some of the absorption above 440 mμ originates from this.

Roth and Schuster have thoroughly tested a number of amino-acids, etc., with a view to the development of yellow-coloured compounds. The absorption spectra of these have been examined in this Department; a few of them (especially tyrosine, phenylalanine and methionine) showed absorption. The character of the compounds formed is under investigation.

The acid-lignin used has kindly been placed at our disposal by Dr. N. E. Warburg, Køge.

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Antibodies in Ribonucleoproteins

IN many hypotheses concerning the mechanism of the immune response the nucleic acids have been assigned a role in the production of antibodies, although no clear evidence for their participation is yet available. As regards protein synthesis in general, there is reason for believing that the ribonucleoproteins lie in the pathway of this synthetic process[1]. Evidence also exists that certain enzymes are present in the ribonucleoprotein complex[2,3], and it is possible that they represent products of protein synthesis mediated by ribonucleoprotein. The present communication describes an attempt to ascertain whether antibodies also are present in ribonucleoprotein complexes isolated from the antibody-forming organs, spleen and lymph nodes, of the rabbit.

Evidence that ribonucleoprotein may be involved in antibody production has already been presented by Sterzl and Hrubesova[4] and Hrubesova *et al.*[5].

They have shown that infant rabbits, still unable to synthesize antibodies, acquire this ability when injected with crude 'nucleoproteins' obtained from the spleens of immunized adult rabbits. The 'nucleoproteins' used in this important and suggestive work were, however, very heavily contaminated with extraneous protein. Furthermore, it is difficult to distinguish between the contribution of donor and recipient animal in this type of experiment. We have therefore employed more highly purified ribonucleoprotein preparations and have attempted an *in vitro* demonstration of antibodies.

The experiments were carried out in the following way. Immunized rabbits (together with several non-immunized control animals) were killed when they showed a high level of serum antibodies. The spleens and sometimes the lymph nodes were removed and used for the isolation of ribonucleoprotein. It was generally found that these organs were larger and heavier in the immunized animals than in the controls.

The isolation of ribonucleoprotein was carried out in a cold room, and a buffer containing 0·002 *M* magnesium sulphate and 0·001 *M* *tris*(hydroxymethyl)aminomethane, *p*H 7·1, was used throughout. Microsomes were prepared from homogenized tissues by a low- (10,000*g*, 30 min.) and a high- (105,000*g*, 2 hr.) speed centrifugation. The microsomes were disrupted with 0·75 per cent sodium deoxycholate, and ribonucleoprotein particles were isolated by two further cycles of high- and low-speed centrifugation[6]. The ribonucleic acid content of the purified material was determined as described elsewhere[2]. As weight per cent of ribonucleic acid plus protein it amounted to about 30–40 per cent for the spleen ribonucleoprotein and 45–50 per cent for the lymph node particles. In nearly all cases the yield of ribonucleoprotein per unit weight of organ was higher for the immunized than the control animals.

Portions of the ribonucleoprotein were digested with pancreatic ribonuclease (1–3 μgm./ml.) under dialysis against several changes of 0·15 *M* sodium chloride at 37° for 5–6 hr., followed by an overnight dialysis in the cold. This treatment removed virtually all the ribonucleic acid. Although the bulk of the protein coagulated, a certain proportion remained soluble. After removal of the coagulum by centrifugation, the soluble protein derived from the degraded nucleoprotein was tested for the presence of antibodies. A number of such tests were also made with undegraded ribonucleoproteins.

In most of our experiments the antigen was sheep red blood cells and the tests were made only with spleen ribonucleoprotein. In these cases the animals were immunized with 1 ml., respectively, of a 30 and 15 per cent suspension of red blood cells injected intravenously six days apart. Six days after the second injection the serum antibody titres of these animals and of the controls were determined, the spleens were removed, and ribonucleoprotein was isolated. The serum titre of the immunized animals was generally of the order of 2^{10}. Direct tests for the presence of agglutinins were made by incubating serial dilutions of either the intact ribonucleoprotein or the soluble protein fraction in physiological saline at 37° with 1 per cent suspensions of the antigen.

No trace of agglutinins was detected in the intact ribonucleoprotein, whether from non-immunized or immunized animals. When, however, the soluble protein fraction derived from the degraded ribonucleoprotein of immunized animals was tested, a typical agglutinin reaction was observed, with titres up to 2^4. In contrast, the soluble proteins from the non-immunized controls caused no agglutination. Thus, antibodies appear to have been present in the spleen ribonucleoproteins of the immunized animals. Their immunological reactivity was apparently not expressed until the ribonucleoprotein was degraded, at which time, presumably, the antibody would be liberated from the nucleoprotein complex.

In view of the low agglutinin titres found in the direct tests, the soluble protein fractions were also subjected to the more sensitive Coombs test. Here, 3 per cent suspensions of sheep erythrocytes (in 0·14 *M* sodium chloride buffered at *p*H 7·1 with 10^{-3} *M* *tris*(hydroxymethyl)aminomethane) were incubated with the soluble proteins for 90 min. at 37°, were washed with buffered saline, and were tested against two times serial dilutions of sheep anti-rabbit serum ('Wellcome' sheep precipitating serum). Those erythrocytes which had been incubated with the soluble ribonucleoprotein proteins of immunized animals were completely agglutinated, being shown thereby to be coated with rabbit antibodies. The Coombs reaction gave a titre of 2^9, with a pro-zone of 2–3 dilutions. Again, the non-immunized controls gave negative tests.

In another experiment non-agglutinating antibodies were tested for. Horse serum globulins, mixed with Freund adjuvant, were injected subcutaneously into rabbits. Nine days later, when the serum titre >3125, spleen and lymph node (inguinal, axillar and brachial) ribonucleoproteins were prepared and degraded with ribonuclease. As noted above, the yield of ribonucleoprotein per unit weight of organ was markedly greater for the immunized than for the non-immunized animals (1·09 vs. 0·53 mgm./gm. of lymph node tissue). Direct interfacial ring tests of the soluble protein fractions gave negative results. Tests were then made with Stavitsky's modification[7] of Boyden's method, wherein the antibody agglutinates antigen-coated sheep erythrocytes (tanned at 37°, sensitized with antigen by exposure to horse serum at room temperature, washed and suspended in a hundred times diluted complement-inactivated (56°, 30 min.) non-immune rabbit serum).

The results (Table 1) show that antibodies were present in the soluble protein fraction derived from the lymph node ribonucleoprotein of immunized animals. The absence of detectable antibodies in the spleen ribonucleoprotein is of doubtful significance, owing to the possibility that the subcutaneous immunization had caused the nodes to be activated earlier than the spleen and to have undergone a more intense response at the time of organ extirpation.

Although the experiments described here are still in a preliminary stage, they indicate that the process of antibody response involves a marked increase in the ribonucleoprotein content of the antibody-

Table 1. Agglutination by Rabbit Ribonucleoproteins of Tanned Erythrocytes Coated with Antigen*

Dilution of antibody solution	×1	×2	×4	×8	×16
Antibody solutions					
Soluble proteins from ribonucleoprotein of:					
(1) Lymph nodes, non-immunized	−	−	−	−	−
(2) „ „ immunized	+++	++	+	−	−
(3) Spleen, non-immunized	−	−	−	−	−
(4) „ immunized	−	−	−	−	−
Controls					
(5) 100 times diluted antiserum	+++	+++	++	+	−
(6) 100 times diluted normal serum	−	−	−	−	−

* Antigen: horse serum globulins.

forming organ, and that the protein moiety of this ribonucleoprotein contains antibodies which are released in an active form upon the destruction of the ribonucleic acid moiety. These observations are consistent with the concept that the antibody molecule forms part of the ribonucleoprotein complex within the antibody-producing cell, and that this complex may, therefore, be the site of its synthesis. Similar observations have been made in the case of the release of the enzymes ribonuclease and deoxyribonuclease from a bacterial ribonucleoprotein[2], and it is possible that this is a general phenomenon of protein biosynthesis.

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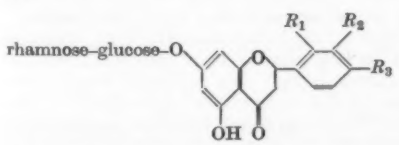
Flavonoids of the Ponderosa Lemon

The isolation and structure of citronin, a new flavanone glycoside from the peel of immature Ponderosa lemons (*Citrus limon* Burm. *f. ponderosa* Hort.) was reported some time ago by Yamamoto and Oshima[1]. From chemical and spectroscopic evidence it was concluded that citronin is 2′-methoxy-5,7-dihydroxyflavanone 7-rhamnoglucoside (*I*) and thus belongs to the rather uncommon group of flavonoids having a substituent at position 2′. In attempting to obtain this compound from samples of both mature and immature Ponderosa lemons, under conditions approximating those of Yamamoto and Oshima, it has been our experience that the only glycoside which can be isolated easily in crystalline form is neohesperidin (*II*). This was identified by elementary analyses, mixed melting point determination, infra-red spectrum, preparation of the phenylhydrazone derivative and isolation of the aglycone hesperetin from the hydrolysate. Neohesperidin differs from hesperidin only in the position of attachment of rhamnose to glucose[2]:



I, $R_1 = OCH_3$; $R_2 = R_3 = H$

II, $R_1 = H$; $R_2 = OH$; $R_3 = OCH_3$

The isolation of neohesperidin from the Ponderosa lemon is of interest for several reasons. First, as Swingle has pointed out[3], "the nature of the characteristic glucoside present in the tissues of a species of *Citrus* may be of definite taxonomic significance in distinguishing that species from another species to which it may have superficial resemblances". The Ponderosa is generally considered to be in the nature of a hybrid which most nearly resembles the lemon (*Citrus limon*), but it is also regarded as having certain characteristics of the citron and grapefruit[4]. Ordinary lemons have not been shown to contain neohesperidin, but do contain hesperidin, diosmin[5] and eriodictyol glycoside[6] as their principal flavonoids. From qualitative tests, we consider that the Ponderosa lemons used in the present work contained little if any hesperidin or eriodictyol glycoside. Therefore, to judge from the identity of the most abundant and easily detectable flavonoids, the Ponderosa lemon would seem to have little relation to ordinary lemons. It is noteworthy that neohesperidin has been found previously only in the sour orange (*Citrus aurantium*)[7] and trifoliate orange (*Poncirus trifoliata*)[8].

Secondly, although it would be unwarranted to suggest that citronin is identical with neohesperidin (since there appear to be several irreconcilable points of difference), it should be noted that some of the chemical evidence reported by Yamamoto and Oshima is incompatible with their structure *I*. According to these authors, citronin (*a*) gives no ferric colour; (*b*) gives *o*-hydrocoumaric acid methyl ether when cleaved with alkali; and (*c*) after hydrolysis to its aglycone gives 5,7,2′-trimethoxyflavanone on treatment with diazomethane. On the contrary, compound *I* or its aglycone would unquestionably give a ferric colour; would be expected to yield *o*-coumaric acid methyl ether (rather than the hydrogenated derivative); and would undergo methylation at the 5-position very slowly, if at all, by means of diazomethane. In fact, some of the evidence suggests that citronin might conceivably be a dihydrochalcone similar to phloridzin.

Finally, it should be mentioned that botanical variations might account for the difference in the results of Yamamoto and Oshima and ourselves. Dr. W. P. Bitters has pointed out to us that there are two forms of the Ponderosa lemon, one of American origin (used in the present work) and the other of Javanese origin, which are undoubtedly of slightly different genotypes but of very similar phenotypes.

We should like to thank Dr. Bitters and Dr. W. B. Sinclair of the Citrus Experiment Station, University of California, Riverside, for their kindness in providing us with authentic Ponderosa lemons.

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An Elastin Gel

The present report describes the conversion of insoluble aorta elastin to a gel. Wood[1] recently described the preparation of an elastin gel from a soluble protein derived from cattle ligamentum nuchæ. These observations may have some bearing on the relationship between collagen and elastin[2–5].

Elastin was prepared from cattle aorta by a method described previously[6], in which 1 *N* sodium hydroxide is used for removing collagen. (Some cattle aorta do not yield insoluble elastin under these severe conditions, perhaps owing to their age.) For the preparation of the gel, the elastin was suspended at room temperature in a 0·1 per cent solution of elastase, in acetate buffer at *p*H 4·7 and ionic strength 0·1. (Two different elastase preparations were used: a lyophilized water extract of porcine pancreatin and porcine elastase *PI*[6].) After at least 10 min., the suspension was centrifuged and the residue washed with the buffer. The washed residue was then suspended in 10 vol. of 0·2 *N* acetic acid, from which, on standing, the gel formed.

Gel formation does not occur when (*a*) cattle ligamentum nuchæ is the elastin source; (*b*) cattle aorta elastin is boiled in 0·1 *N* sodium hydroxide; or (*c*) the enzyme treatment is omitted.

The gel melts upon either warming or adding sodium chloride, with subsequent formation of an elastin precipitate. An approximation of the melting point can be made by suspending 1 vol. of packed gel in 5 vol. of aqueous solvent and noting the temperature at which flocculation begins. As the concentration of sodium chloride of the suspension medium increases from 0·01 *M* to 2·0 *M*, the melting and precipitation point first rises and then falls, the maximum occurring between 0·05 and 0·5 *M* sodium chloride. Syneresis has never been observed, even after prolonged storage at 4°C., in the absence of salt.

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PHYSIOLOGY

Experimentally Induced Proliferation of the Rudimentary Gonad of an Intact Domestic Fowl

It is well known that the right female gonad of the domestic fowl ceases to grow about the ninth day of incubation and that it always hypertrophies following sinistral ovariectomy. I have shown[1] that œstrogen could act *in vivo* as the inhibitor of rudiment growth. In view of the anti-œstrogenic effect of 17 α-ethyl-19-nortestosterone ('Nilevar', Searle) upon the chicken's oviduct[2], the effect of this substance upon the rudimentary chicken gonad was investigated. Starting at 60 days of age intact White Leghorn females received 40 subcutaneous injections of 'Nilevar'. The substance was dissolved in a carrier of 5 per cent benzyl alcohol in corn oil and 0·1 c.c. of solution was injected daily. Experimental procedures were analogous to those outlined in the earlier paper[1] and the weights of blotted, fixed (Bouin's fixative) tissue were recorded (Table 1).

Table 1

No. of birds	Treatment	Rudiment weight (mgm.) at 100 days ± standard error of the mean
9	Carrier	5 ± 1
10	1·5 mgm. 'Nilevar'/day	13 ± 1
8	3·0 mgm. 'Nilevar'/day	20 ± 2

Statistical analysis showed the differences due to treatment to be highly significant.

This is the first report of experimentally induced proliferation of the rudimentary intact fowl gonad. Detailed results, including evidence that the proliferation is due to the anti-œstrogenic action of 'Nilevar', will be published elsewhere.

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Effect of Testosterone Propionate and Insulin on Muscle Cholinesterase Activity

Various hormones influence the cholinesterase activity of tissues[1,2]. The purpose of the present work was to investigate the influence of some anabolic hormones such as testosterone and insulin on muscle cholinesterase activity.

Muscle cholinesterase was studied on male albino rats weighing 250 gm. and kept on a standard stock food. The animals were divided into three groups. One group received 1 mgm. testosterone propionate in 0·2 c.c. sesame oil per animal for ten days, the second group 2 I.U. of crystalline insulin per kgm. body-weight for the same period. The third group was used as control. The animals were autopsied 24 hr. after the last injection, and the cholinesterase activity was determined in diaphragm and gastrocnemius in tissue homogenates by Michel's method[3]. The results (Table 1) are expressed in Δ *p*H/hour per gm. of wet tissue weight.

Table 1. Effect of Testosterone Propionate and Insulin on Rat's Gastrocnemius and Diaphragm Cholinesterase

	No. of animals	Gastrocnemius	Diaphragm
Control	24	0·783 ± 0·068	0·910 ± 0·070
Testosterone propionate (1 mgm. per day)	12	1·410 ± 0·110 ($P < 0·01$)	1·065 ± 0·050 ($P < 0·05$)
Insulin (2 I.U. per kgm. body-weight per day).	12	1·157 ± 0·048 ($P < 0·01$)	1·204 ± 0·064 ($P < 0·05$)

The cholinesterase activity is expressed in Δ *p*H/hour per gm. of wet tissue. The determinations were carried out on 0·2 gm. wet tissue.

As is seen in Table 1, testosterone propionate produces a significant increase in cholinesterase activity of gastrocnemius ($P < 0·01$) and a rather moderate increase in diaphragm cholinesterase activity

($P < 0.05$). Insulin has virtually the same effect (Table 1). An increase in muscle cholinesterase activity is also observed during embryonic development where marked anabolism occurs[4]. Therefore a relationship is suggested between anabolism and cholinesterase activity in striated muscles. The above results seem to support such a hypothesis since testosterone has a well-known myotropic effect[5] and insulin is also considered as an anabolic agent[6].

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Assaying Luteinizing Hormone

There is a demand for an easy and accurate method of assaying the luteinizing fraction of gonadotrophic hormone. The present method depends upon the formation of corpora hæmorrhagica in the ovaries of immature female mice after rendering their Graafian follicles mature. The gonadotrophic materials used were pregnant mare serum ('Antex', Leo) as a source of follicle stimulating hormone, and chorionic gonadotrophin ('Physex', Leo) as a source of luteinizing hormone. The material used for assay was dissolved immediately before use in sterile saline and administered as a single dose to avoid deterioration of the hormone by storage even when refrigerated[1].

Table 1. Influence of Dose of Pregnant Mare Serum on the Corpus Hæmorrhagicum Response

Dose of PMS (I.U.)	Dose of HCG (I.U.)	No. of animals	Average No. of corpora hæmorrhagica/mouse*
20	None	15	0·20 ± 0·03
20	10	15	3·66 ± 0·26
40	None	15	0·28 ± 0·07
40	10	15	3·86 ± 0·10
80	None	15	0·30 ± 0·03
80	10	15	3·43 ± 0·36

* ± Standard error.

In order to determine the dose of pregnant mare serum and the time after its administration when the Graafian follicles become fully developed and ready for ovulation, 90 immature female Swiss mice, 3–4 weeks old, were used. They were divided into 3 groups which were injected subcutaneously with a single dose of either 20, 40, or 80 I.U. of pregnant mare serum. The number of animals with opened vaginas were counted daily. On the fourth day there were more than 80 per cent of these. Half the animals of each group were injected subcutaneously with a single dose of 10 I.U. of chorionic gonadotrophin and vaginal smears were taken daily. Forty-eight hours after administration, all smears were either metœstrus or dicestrus. The animals were killed with ether, their ovaries dissected, and the number of corpora hæmorrhagica formed in both ovaries of each animal was recorded.

After establishing the time relations of the assay method, 4 runs of assay were planned. Two assays were run together. In one case the initial dose of pregnant mare serum was 40 I.U. and in the other 20 I.U. After four days the animals were given graded doses of chorionic gonadotrophin as shown in Table 2. The third assay was planned to determine if variations in the follicle-stimulating hormone/luteinizing hormone ratio would influence the slope of the regression line and if it induces a source of error in the assay. 4 days after the injection of 20 I.U. of pregnant mare serum as usual, a total dose of 25 I.U. of different combinations of pregnant mare serum and chorionic gonadotrophin were injected as presented in Table 3. The assay

Table 2. Corpus Hæmorrhagicum Response to Graded Doses of Chorionic Gonadotrophin

Dose of HCG (I.U.)	Average No. of corpora hæmorrhagica 40 I.U. PMS	20 I.U. PMS*
None	0·28 — 0·07	0·33 ± 0·10
1	2·86 — 0·09	3·28 ± 0·33
10	4·86 — 0·48	5·83 ± 0·28
100	8·03 — 0·85	9·83 ± 0·84
1,000	—	9·25 ± 0·98

* ± Standard error. Each group contained 7 mice.

Table 3. Influence of Follicle Stimulating Hormone/Luteinizing Hormone Ratio on Corpus Hæmorrhagicum Response

Dose of PMS (I.U.)	Dose of HCG (I.U.)	Average No. of corpora hæmorrhagica
None	None	0·33 ± 0·05*
5	20	6·00 ± 0·28
10	15	5·00 ± 0·33
15	10	4·50 ± 0·38
20	5	3·80 ± 0·36

* ± Standard error.
Each group contained 7 animals.
All mice were previously treated with 20 I.U. of PMS.

No. 4 was performed to test if blood serum could modify or augment the corpus hæmorrhagicum response to luteinizing hormone as happens in other methods[2]. A comparison was made between the corpus hæmorrhagicum response to 10 I.U. of chorionic gonadotrophin dissolved in saline with that dissolved in ox serum (Table 4).

Table 4. Influence of Addition of Serum on the Corpus Hæmorrhagicum Response

Treatment	Average No. of corpora hæmorrhagica*
0·5 ml. ox serum	1·25 ± 0·12
10 I.U. of HCG + 0·5 ml. ox serum	5·00 ± 0·86
10 I.U. of HCG + 0·5 ml. saline ..	4·75 ± 0·12

* ± Standard error.
Each group contained 8 animals.
The animals were previously treated with 20 I.U. PMS.

The average of the number of corpora hæmorrhagica formed in an animal was plotted against the logarithm of the dose. This dose response curve showed a straight line, up to 100 I.U. of chorionic gonadotrophin. The slopes of the response curves constructed under these conditions were estimated and tests of the hypothesis of equality of the regression lines were performed. The results presented in Tables 1 and 2 show that the initial dose of follicle-stimulating hormone did not modify the corpus hæmorrhagicum response to luteinizing hormone administration. When 20 or 40 I.U. of pregnant mare serum were used, the slopes of the two regression lines were not different ($t = 0.19$). The results presented in Table 3 show that variations in follicle-stimulating hormone/luteinizing hormone ratio of the test material had no influence on the number of corpora hæmorrhagica formed by luteinizing hormone. It appears also that ox serum did not affect the ovarian response to luteinizing hormone. The passage of the ovaries through susa, alcoholic iodine, alcohol, methyl benzoate and finally benzene renders them transparent and corpora hæmorrhagica could be easily counted.

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Carbon Dioxide Assimilation by *Leishmania donovani*

While studying the glucose metabolism of *Leishmania donovani*, the protozoan organism causing visceral leishmaniasis, it has been found that cell suspensions of this organism in a saline-bicarbonate buffer containing glucose can assimilate a measurable amount of carbon dioxide along with the formation of different acidic end products[1].

L. donovani, strain 81, cultured on Ray's medium[2] at 22–24° C. and recycled every three weeks from the Nicolle-Novy-McNeal medium, was used in this study. After growth for three days on Ray's medium, the organism was scraped from the surface of the agar, washed twice with ice-cold isotonic saline by centrifugation at 3,000 r.p.m. for 10 min. and finally resuspended in isotonic saline.

Standard conditions used for the measurement of carbon dioxide assimilation were as follows: 3·0 ml. of the complete reaction mixture contained 15 μ moles of sodium bicarbonate (*p*H 7·3), 30 μ moles of glucose, 1 ml. of cell suspensions in isotonic saline containing 1·5–2 mgm. protein nitrogen. Incubations were carried out for one hour in conventional Warburg manometric vessels at 37·2° C. in an atmosphere 95 per cent nitrogen and 5 per cent carbon dioxide.

Results from three typical experiments showing the carbon dioxide balances during anaerobic glucose breakdown by cell suspensions of *L. donovani* are presented in Table 1. It will be seen that the amount of carbon dioxide assimilated by the organisms under these conditions varies from 32 μl. to 80 μl. The variation in the amount of carbon dioxide fixation was afterwards found to be due to the different growth stages of the culture used. Table 2 represents figures for the stoichiometric relationship between glucose utilized, acid produced and carbon dioxide fixed in three typical experiments carried out under standard experimental conditions. It will be seen that nearly 0·30–0·36 mole of carbon dioxide is incorporated per mole of glucose consumed. Results using growing culture indicate that the amount of carbon dioxide fixed varies with the age of culture; with two days old growing culture the molar ratio of carbon dioxide fixed per mole of glucose utilized is nearly one. From Table 2 it will also be seen that an average of 1·5 moles of acids are formed per mole of glucose metabolized. Chemical analysis of the different acids that are formed indicated that the major products were non-volatile in nature, and among them succinic acid predominated although malic acid formation has also been noted.

Table 1. Carbon Dioxide Balances during Anaerobic Breakdown of Glucose by *L. donovani* Suspensions

No. of Exp.	Total available carbon dioxide (μlitre)	Carbon dioxide liberated due to acid formation	Residual carbon dioxide	Carbon dioxide fixed
	a	*b*	*c*	*a*–(*b* + *c*)
1	262	144	74	44
2	267	150	37	80
3	126	82	12	32

Table 2. Carbon Dioxide Assimilation by *L. donovani* Suspensions

Expt. No.	Glucose consumed (μmole)	Total acid formed (μmole)	Carbon dioxide fixed (μmole)	Carbon dioxide fixed/ Glucose used
1	3·1	4·4	1·1	0·36
2	6·1	8·6	1·9	0·31
3	5·3	7·8	1·6	0·30

When glucose was replaced by glycerol, ribose or pyruvate, no measurable amount of carbon dioxide fixation was noticed. Of the various inhibitors tried, DNP (10^{-4} *M*), α-α′-dipyridyl (10^{-3} *M*) and 8-hydroxyquinoline (10^{-3} *M*) were found to have marked inhibitory effects on the glucose utilization as well as on the carbon dioxide assimilation. It seems that the carbon dioxide assimilation by this protozoal organism is in some way related to the glucose metabolism. The high rate of carbon dioxide assimilation may also be complementary to the very active transaminase system reported to be present in this organism[3].

Detailed studies on the biochemical basis for the carbon dioxide requirement of this protozoal organism are in progress and will be reported elsewhere.

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HÆMATOLOGY

Hæmolytic Properties of Immune Chicken Antibody

In the course of a study of immunological responsiveness of chickens to foreign red blood cells our interest was attracted by the observed discrepancy between high agglutination titres of immune chicken sera and their inability to show lytic properties in the presence of complement. It is well known that different antibody–antigen systems exhibit different abilities to fix guinea pig complement when mammals are used as antibody producers[1]. The failure of immune bird sera to show a significant complement-fixing activity was mentioned by Meyer[2] and by Rice[3] in birds infected with ornithosis virus and *S. pullorum*. It was postulated that avian antibody formed with antigen non-complement-fixing aggregates[4], and that avian antibody lacked some character that was important for combining with complement[5]. This problem, however, was not further explored, and all available data were based on the experiments made with guinea pig complement. It seemed, therefore, reasonable to investigate in a preliminary way whether or not chicken antibody hæmolyses red cells in the presence of complement from different animal species.

Hæmolytic sera were prepared by inoculating intravenously Rhode Island Red pullets with 5 per cent suspension of duck and rabbit erythrocytes (two series of three injections each, followed by re-injection after 20 days). The diluent for all reagents was sodium chloride–veronal buffer *p*H 7·3–7·4 containing

Table 1. HÆMOLYTIC ACTIVITY OF NORMAL ANIMAL AND IMMUNE AVIAN SERA IN THE PRESENCE OF COMPLEMENT FROM VARIOUS SOURCES

Normal animal serum				Immune chicken serum (hæmolysins)	
Species (source of complement for immune hæmolysis)	Complement units in 1 ml.	Hæmolysins for duck red cells	Hæmolysins for rabbit red cells	anti-duck*	anti-rabbit*
Chicken	10·41	–	+1 : 25	–	×
Duck	24·34	–	+1 : 32	–	×
Owl	3·03	–	+1 : 10	–	×
Swan	31·40	–	+1 : 20	–	×
Eagle	32·47	–	+1 : 32	–	×
Wild goose	30·0	–	+1 : 32	–	×
Pelican	34·70	–	+1 : 32	–	×
Ground squirrel	<1·0	–	–	–	–
Guinea pig	180·50	–	–	–	–
Rat	13·45	–	–	–	–
Rabbit	24·45	–	–	–	–
Dog	55·52	+1 : 25	–	×	–
Cat	6·63	+1 : 32	+1 : 16	×	×
Horse	<1·0	–	–	–	–
Ox	2·49			–	–
Sheep	<1·0	–	–	–	–
Pig	18·21	+1 : 2	+1 : 2	–	–
Rhesus monkey	66·45	+1 : 40	+1 : 40	×	×
Man	32·38	+1 : 32	+1 : 32	×	×
Frog	56·40	+1 : 8	+1 : 32	–	×

–, No hæmolysis; +, hæmolysis; ×, the hæmolytic reaction was not done because of the presence of normal hæmolysins in the quantity of the serum required as complement source.
* In all cases the complement units were present in the supernatants after incubation.

optimal concentrations of calcium and magnesium ions[6]. Sheep, duck and rabbit red cells (3 per cent suspension; *O.D.* 0·453; Beckman spectrophotometer at 541 mμ) were standardized as in Osler *et al.*[7] Pools of fresh sera from different animal species served as the source of complement. The sera were stored at – 65° C. The hæmolytic complement activity of pools was titrated with sheep red cells sensitized with rabbit hæmolysin, and the titre estimated in 50 per cent hæmolytic units. Two and five 50 per cent complement units were used in titration of chicken antisera. The dilutions of antisera were in the range of 1 : 2 to 1 : 3,000. Duck and rabbit erythrocytes were sensitized in chicken anti-duck and anti-rabbit sera, respectively, for 30–90 min. The incubation with two or five complement units lasted 60–90 min. All reactions were carried out at 37° C. Complement hæmolytic units left in supernatants were estimated with sensitized sheep erythrocytes (rabbit hæmolysin). The chicken immune sera were also examined for the presence of agglutinins, and pools of animal sera were investigated for the presence of normal hæmolysins for duck and rabbit erythrocytes.

Agglutination titres of immune chicken sera were 1 : 1,000–1 : 4,000 for rabbit, and 1 : 120–1 : 1,000 for duck red cells. Some animal sera were very poor in complement (ground-squirrel, horse, sheep, ox and owl). It is evident that complement of different animal species was involved in the hæmolysis of sensitized sheep erythrocytes. On the other hand, no hæmolytic activity of chicken anti-duck and anti-rabbit sera could be observed in the presence of complement. It is interesting to note that normal hæmolysins in avian and animal sera exhibit hæmolysis. This observation may suggest that some differences exist between normal hæmolysins and immune hæmolysins in avian sera.

It is difficult to believe that avian species are not able to produce immune hæmolysins. The failure of immune chicken antibody to hæmolyse red blood cells is probably due to its peculiar behaviour in fixing complement. It should, however, be borne in mind that the complement-fixation is a multiple step reaction of very complex nature which may be affected by a variety of conditions, and in which the complement components react disproportionately[8]. Our knowledge about complement reaction is based mainly on the data obtained from the experiments with guinea pig complement and sensitized sheep erythrocytes. But one is confronted with a somewhat different situation when working with avian antibody. The present experiments suggest that immune hæmolysis in which avian antibody is involved differs to some extent from other immune hæmolyses and requires further study which will probably reveal important information about the mechanism of the action of complement.

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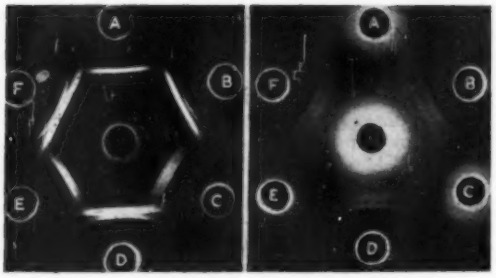
Individual Specific Nature of the Cold Auto-Antibodies of Acquired Hæmolytic Anæmia

DURING the course of an immuno-chemical study of the cold agglutinins present in the sera of patients suffering from acquired hæmolytic anæmia of the cold-antibody type, evidence was obtained which indicated that they were immunologically distinct.

Sera from seven patients were available. Antisera were prepared in rabbits, (1) against the cold antibodies of two of these patients, eluted from erythrocyte stroma, (2) against separated cryoglobulin (acting as a cold antibody) derived from one patient, (3) against γ_1-globulin separated by starch-block electrophoresis from the serum of a patient with acquired hæmolytic anæmia secondary to lymphosarcoma, and (4) against γ_1-globulin separated from a normal serum. The antisera were tested for their reactions by double diffusion in agar gel and immuno-electrophoretic techniques[1].

When the rabbit antisera produced against the pathological cold agglutinins were absorbed with 2·5 mgm. per ml. of γ-globulin (Lister Institute, London) and tested by double diffusion in agar gel against the homologous and heterologous cold-antibody-containing sera and a normal serum, the normal serum produced a single line of precipitate whereas each of the pathological sera produced two lines of precipitate (Fig. 1*a*). The lines formed by the homologous serum showed a reaction of partial identity (cross-reaction) with those formed by the heterologous sera or normal serum, as indicated by spur formation. This suggested the presence of individually specific groupings in the homologous cold-agglutinin protein.



a *b*

Fig. 1. *a*, Photograph of an agar-gel-diffusion plate showing the precipitation pattern of a human serum containing high-titre cold agglutinin (cups *B*, *D* and *F*) and two other human sera also containing high-titre cold agglutinins (cups *C* and *E*) and a normal human serum (cup *A*). The central cup contains rabbit antiserum against the cold agglutinin of the serum used in cups *B*, *D* and *F* and was absorbed with a minimal amount of γ-globulin. The human sera were diluted 1 in 4. *b*, Photograph of an agar-gel-diffusion plate showing the precipitation pattern of a human serum containing high-titre cold agglutinin (cups *B*, *D* and *F*) and two other human sera also containing high-titre cold agglutinins (cups *C* and *E*) and a normal human serum (cup *A*). The central cup contains rabbit antiserum against the cold agglutinin of the serum used in cups *B*, *D* and *F* and was absorbed with twice its volume of normal serum (central cup). The human sera were diluted 1 in 4

When the antisera were further absorbed with an excess of pooled normal human serum or with the γ_2- and γ_1-globulin fractions separated electrophoretically from normal human serum, they reacted with homologous serum only (Fig. 1*b*). No reaction was discernible with the heterologous pathological sera or normal human serum even when the gel-diffusion plates had been kept at 37° C. for 3 months. Exactly similar reactions were obtained when the cold agglutinins eluted from erythrocyte stroma were used as test antigens in place of the pathological sera.

Further experiments showed that the formation of precipitin lines by the homologous serum only could not be explained by non-specific precipitation due to the presence of heat-labile components of complement, by incomplete absorption of the antisera or by the presence of different globulin groups[2] in the serum used for absorption and the patient's serum. It is of interest to note that when the rabbit antiserum produced against a normal γ_1-globulin was similarly absorbed, it failed to react with any normal or pathological serum, indicating complete neutralization of the antibodies contained in it.

Although specific globulins have been identified by their Gm groups[2] and specific differences reported between the macroglobulins of Waldenström's macroglobulinæmia[3], globulins with antibody properties are not usually considered as having individually specific characteristics. The immunological differences observed may have a counterpart in the subtle differences in serological properties already observed by Dacie and de Gruchy[4] in the antibodies of acquired hæmolytic anæmia. The presence of antigenic groupings specific to the cold antibodies of each individual is in accord with the hypothesis that the formation of cold antibodies represents the synthesis of abnormal proteins rather than an immune response on the part of normal antibody-forming tissues. The observation could perhaps be taken to favour the concept of somatic mutation occurring in the antibody-forming tissues of the patient.

I am indebted to Dr. J. H. Humphrey, of the National Institute for Medical Research, Mill Hill, London, for advice and helpful criticisms and for the supply of γ-globulin preparations used in this investigation.

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Distribution of Gm[a] and Gm-like among Javanese, Djuka Negroes, and Oyana and Carib Indians

Grubb and Laurell[1] and others have demonstrated that some human sera contain a genetically determined factor capable of inhibiting the ability of serum from selected rheumatoid arthritic patients (RA serum) to agglutinate Rh(+) cells coated with selected incomplete anti-D sera. They called this factor Gm[a] and showed that its presence is due to a dominant gene. Thus far various investigators have tested 2,635 Europeans and found that 56·96 per cent were Gm(a+)[2]. Grubb and Laurell[1] found 95 per cent of 74 Eskimos to be Gm(a+), and Steinberg and Giles[2] found 98 per cent of 98 American Negroes to be Gm(a+).

Steinberg and Giles[2] found an RA–anti-D system which tested for a factor which is not Gm[a], Gm[x][2], or Gm[b] (ref. 4 and Harboe, M., personal communication). Because its genetic relation to these factors is unknown, the new factor is tentatively called Gm-like. This factor was present in 27·5 per cent of 403 American Negroes and absent in all of 250 Whites. Its presence is determined by a dominant allele.

We have now tested serum from 20 Javanese, 35 Djuka Negroes, 15 Oyana Indians, and 19 Carib Indians for Gm[a] and Gm-like. The method of testing was that described by Grubb and Laurell[1]. The samples were collected by Dr. Vincent Butler in Surinam (Netherlands Guiana). The Djuka Negroes are descendants of slaves who escaped into the jungle shortly after importation into Dutch Guiana. The Oyana Indians are an isolate of jungle Indians, living along a river in the interior of Surinam. The Carib Indians are coastal. So far as we know, these people have not intermarried with other ethnic groups.

All 89 samples were positive for Gm[a]. The tests for Gm-like were done blind in the sense that all sera were numbered with no indication of the ethnic origin of their donors. The results of the tests for Gm-like are presented in Table 1.

The samples are small; hence, it cannot be established that the difference in frequency of Gm-like(+) among Javanese and Djuka Negroes is significant ($P > 0{\cdot}3$), if indeed the difference is

Table 1. Results of Tests for Gm-like on Sera from Javanese, Djuka Negroes, Oyana Indians and Carib Indians

	Total	Gm-like + No.	Gm-like + Percentage	Gm-like − No.	Gm-like − Percentage
Javanese	20	10	50	10	50
Djuka Negroes	35	22	63	13	37
Oyana Indians	15	0	—	15	100
Carib Indians	19	0	—	19	100

significant. There is little doubt, however, that the frequency of Gm-like(+) among the Indians is less than that among the other two groups [P (Javanese–Oyana) $\sim 0{\cdot}002$, P (Djuka Negroes–Oyana) $< 0{\cdot}0001$]. Similarly, there is little doubt that the frequency of Gm-like(+) among the Indians is less than that among American Negroes [$\chi^2_{(1)}$ (corrected) $= 4{\cdot}33$, $P < 0{\cdot}05$ for Oyana versus American Negroes] or that the frequency of Gm-like(+) among the Djuka Negroes is greater than that among the American Negro ($\chi^2_1 = 19{\cdot}79$, $P < 0{\cdot}001$).

The data are too few to permit a discussion of the ethnic and racial distribution of these genes (*Gm*^{a} and *Gm-like*) beyond stating that the frequency of *Gm*^{a} among the coloured peoples seems to be 100 per cent and that *Gm-like* is essentially absent in Whites and possibly in South American Indians and occurs among Javanese and Negroes. Further data are required. The wide variation in frequency of *Gm-like* among various ethnic groups indicates that it may be a useful tool for anthropologists and others interested in population problems.

We are deeply indebted to Dr. Butler for making these sera available to us and to the hospital and laboratory of the Aluminium Co. of America for making their facilities in Surinam available for the collection of the blood specimens used in this study.

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Absence of the Diego Blood-Group Antigen in the Lapps

Blood-group studies in the Lapps have revealed rather unusual distributions within several blood-group systems[1,2]. The Lapps seemed to have a curious mixture in their blood groups of European features on one hand, and Asiatic ones on the other.

The discovery of the Diego blood-group antigen has given a valuable tool in anthropological investigations[3]. As the Diego antigen is confined to Mongoloid populations, and in view of the possibility of the Lapps being related to these populations, it seemed to be of importance anthropologically to determine whether this factor was present in the Lapps.

We have tested a total of 433 blood samples from Norwegian Lapps for the Diego factor. The anti-Di^a serum and the Di(a+) control cells used in the study were generously placed at our disposal by Dr. Miguel Layrisse, of Caracas, Venezuela. The serum reacted strongly by the indirect Coombs's technique against the Di(a+) cells, whereas all the 433 Lapp blood samples gave negative results.

From Sweden, further results on the blood groups of the Swedish Lapps have just been published[4]. These investigations also include Diego grouping on 220 blood samples, all of them giving negative reactions. Pooling the Swedish and Norwegian data, we find that a total of 653 Lapps have been examined for the Di^a factor. The absence of the factor in a sample of this size seems to exclude any close relationship to the Mongoloid populations carrying this antigen. In this connexion we should, however, like to point out that the Diego antigen was also absent in a population which is usually regarded as Mongoloid, namely, Eskimos from the eastern Canadian Arctic[5,6].

The Norwegian Lapp blood samples were also grouped in the A_1A_2BO, MNSs, P, Rh(C,c,C^w,D,E,e,V), Kell, Lewis(Le^a,Le^b), Duffy(Fy^a), Kidd(Jk^a,Jk^b), and Wright blood-group systems. A report of these investigations will be given elsewhere.

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Haptoglobin in Monkeys

We have examined the haptoglobin types of a series of blood samples from 27 monkeys (23 *Macaca mulatta*, 1 *Macaca irus* and 3 *Cebus nigrivittatus*). The *Macaca mulatta* and *Macaca irus*, indigenous to India and Java, were purchased from the United States (Trefflich's Bird and Animal Co. Inc.). Since the rhesus monkey species infrequently reproduces itself in captivity, it can be assumed, even though no genetical results were obtained, that these monkeys are probably unrelated. The few *Cebus* samples examined came from the Higuerote (Miranda State) and Tamanaco (Guárico State) regions, in Venezuela.

The method used for the study of haptoglobins and their differentiation is based mainly on the methods of Smithies[1] and Allison and ap Rees[2] and has already been outlined[3]. Thanks to these methods there exists no doubt as to the classification of haptoglobins.

All the monkeys examined, both from Venezuela and from India and Java, were found to have type 1–1 haptoglobin, probable genotype $H_p{}^1/H_p{}^1$.

The number of monkeys used in our studies is relatively small for definite conclusions to be reached. Should future investigation confirm that monkeys have only one type of haptoglobin, then this finding could be phylogenetically significant, since both monkeys and man belong to the same primate order. In all probability there only existed in the past what is now called type 1–1 haptoglobin, from which the other types known at present are derived by mutation. It would be interesting to know, in this connexion, whether any human populations exist the haptoglobin of which belongs to type 1–1 only. If not, one would be inclined to think that this change probably occurred during or after the evolution of the *Homo sapiens* from non-human and early human types. It

is not yet known whether such a mutation has or has not actually favoured this evolution.

Several authors[4–6] have dealt with the much discussed problem of whether the polymorphism of present man is either a recent acquisition or has been directly inherited from his non-human ancestors. Studies made on several species of monkeys in relation to their blood groups as well as to their ability to taste phenylthiocarbamide[4,7] have revealed that the genetical polymorphism of man, in this connexion, is a property common to all primates. If our findings are confirmed, we may be facing a gene (H_p) specifically showing a difference between man and other primates.

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A Survey of Abnormal Hæmoglobins in Portuguese Guinea

In 1957–58 we conducted a survey of abnormal hæmaglobins among the indigenous population of Portuguese Guinea, during which 9,224 blood samples were collected.

In each case, erythrocytes were tested for sickling, and their hæmoglobin was examined by paper electrophoresis of *p*H 8·6. When fast-moving hæmoglobins were detected these were submitted also to paper electrophoresis at *p*H 6·5 and compared with standard samples of known hæmoglobin variants. In the case of hæmoglobin *N*, the first sample we collected was kindly classified by Dr. H. Lehmann in London; after this, further instances of hæmoglobin *N* were classified by ourselves by comparison with this first sample. For identification of hæmoglobin *K*, a sample of *K* obtained during a previous survey at St. Thomas and Prince Island, and one of hæmoglobin *J* supplied by Dr. Lehmann were used. The results of the survey are given in Table 1.

The abnormal hæmoblogins found were *S*, *C*, *K* and *N*.

Not infrequently we found a blood to give a positive sickling test, but not to show the presence of hæmoglobin *S* on electrophoresis. In every case, blood was again withdrawn and submitted to the same tests. In every case, hæmoglobin *S* proved once more to be absent on paper electrophoresis, and the sickling test was negative on the second occasion. We consider that sickling-rates among populations based solely on the sickling test are subject to error, due to false sickling.

The frequency of hæmoglobin *S* varies greatly among the different tribes. As a rule we can say that it is much more common among the Moslem tribes than among the Fetichists.

Hæmoglobin *C* was seen only among the Moslem tribes, which are those with closer relationships with the neighbouring countries.

Hæmoglobin *K* was first described by Cabannes and Buhr[1] among the Algerian Kabyles. Later, Robinson *et al.*[2] found it twice in Liberia, calling it hæmoglobin Liberian II. It has been further found by Ager and Lehmann[3] in an East Indian family, and by Edington and Lehmann among 3 Dagombas of the Northern Ghana, by Cabannes and Sansarricq[1] among 2 Senefous of the High Volta, by Cabannes and Gavanou[1] in one Touareg from the Hoggar, and by Trincão *et al.*[4] in 2 individuals from St. Thomas Island. In Portuguese Guinea we found 8 instances of this rare hæmoglobin; 6 among the Moslem tribes, and 2 among the Fetichists.

Table 1

Tribes	Total No. of observations	Hb. *AS*		Hb. *AC*		Hb. *AK*		Hb. *AN*	
		No. of cases	Per cent	No. of cases	Per cent	No. of cases	Per cent	No. of cases	Per cent
Bagas* †	8					1			
Balotes	342	1	0·2						
Balantes	776	7	0·9			1	0·12	1	0·12
Banhuns	63	10	15·8						
Beafadas†	797	103	12·9	1	0·13	1	0·13	1	0·13
Bijagos	632	2	0·47			1	0·15		
Cassangas†	144	34	23·6						
Felupes	641	36	5·5						
Fulas	1,553	248	15·9	6	0·4	2	0·13	4	0·25
Jacancas* †	43	4	9·3						
Jalofos* †	1								
Landoman* †	3								
Laibe* †	3								
Mancanha	534	1	0·2			2	0·4	1	0·2
Mandinga†	895	115	12·8	10	1·12	1	0·1		
Manjaco	758	16	2·1			1	0·13	3	0·4
Miefora* †	1								
Nalus†	100	6	6						
Nar* †	2								
Pajadinca†	300	48	16			1	0·33	2	0·66
Papel	1,244	16	1·3						
Saracole	268	44	16·4	4	1·5				
Sosso†	94	3	3·1					2	2·1
Surua* †	10	2							
Tilbonca* †	8	3							
Timene* †	4								

* Tribes living in neighbouring territories and encountered by chance in Portuguese Guinea.
† Moslem tribes.

Hæmoglobin *N* seems to be very rare. It was first seen in 8 members of the Mande tribe of Central Liberia by Robinson *et al.*[2], who called it Liberian I. Ager and Lehmann mention two further instances, one from Ghana and one from Nigeria. Further instances have been reported from Portuguese Guinea. It seems interesting to point out that recent surveys of abnormal hæmoglobins among some 2,000 Nigerians failed to reveal any further example of this rare hæmoglobin variant (personal communication, Dr. H. Lehmann).

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PATHOLOGY

Chorioallantoic Membrane Lesions produced by Inoculation of Adult Fowl Leucocytes

Interest in the reaction of the chick embryo to inoculation with adult homologous tissues, noted in 1916 by Murphy[1], has been renewed by the investigations of Simonsen[2] and Cock and Simonsen[3], which indicate that the alterations in the embryo, including splenomegaly, hepatomegaly, and hæmolytic anæmia, result from an immunological attack by the adult cells on the embryonic tissues. This communication reports that cell suspensions prepared from adult fowl blood, spleen or thymus, when placed on the chorioallantoic membrane, produce characteristic membrane lesions in addition to splenomegaly and hepatomegaly. The chorioallantoic lesions closely resemble the splenic nodules observed by Isacson[4] which developed in 11–15 day chick embryos following intravenous injection of adult homologous leucocytes or spleen cells. Isacson suggested that the splenic nodules represented foci of adult donor cells which were stimulated to proliferate by contact with foreign embryonic antigens, and which were afterwards walled off and destroyed by the maturing embryo.

The chorioallantoic lesions produced by inoculation of adult homologous cell suspensions on to the membrane of 10–14 day old embryos required 4–5 days to develop to the stage illustrated in Fig. 1. At this mature stage they appeared as raised yellow or white nodules, 2–4 mm. in diameter, showing a semi-translucent peripheral zone, and a firm, opaque central core which could be readily shelled out from the surrounding tissue. Multiple tiny opaque areas were sometimes present as well, but they were a less constant feature. Microscopically the inoculated membranes showed a complex variety of changes; only those which appear to be concerned directly with the characteristic nodular lesions will be considered here. The sequence of development of the nodular lesions was similar to that of the splenic nodules described by Isacson[4]. Within 48 hr. after inoculation with adult cells, rounded collections of multiplying mononuclear or spindle-shaped cells were observed in the mesodermal layer of the membrane. By three days, many fibroblasts, mononuclear cells and granulocytes

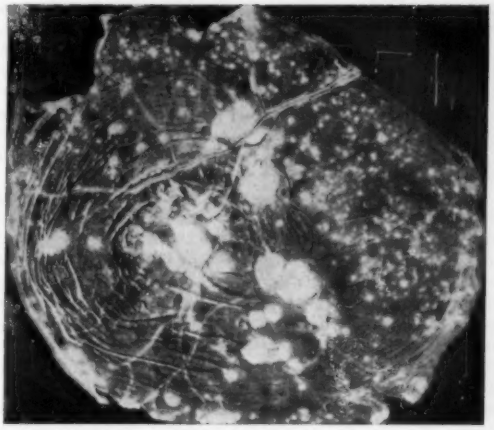

Fig. 1. Chorioallantoic membrane from a chick embryo inoculated at 12 days with a suspension of adult fowl leucocytes and harvested after 6 days incubation at 38° C. Typical large nodules are shown with many small foci which may be of secondary origin.

were found surrounding the cell foci. Somewhat later the foci were invaded by granulocytes and underwent necrosis. Infiltration of the adjacent membrane tissue with granulocytes was a frequent and striking feature in the late stages. The alterations in the chick embryo were similar to those which occurred subsequent to intravenous injection of adult homologous blood and spleen cells[2,4]. Any given cell concentration, however, evoked more marked changes in the embryo when given intravenously than when inoculated on to the chorioallantoic membrane. Following either route of inoculation, characteristic nodules developed in the spleen and less frequently in other sites, including the thymus, proventriculus, bone, muscle and subcutaneous tissue. Widespread granulopoiesis occurred, particularly in the periportal areas of the enlarged liver.

The production of the chorioallantoic and associated embryonic lesions by adult fowl cells was a highly reproducible phenomenon. The blood of all the eight adult fowl donors tested, including four in-bred White Leghorn, one New Hampshire Red and three White Leghorn-Australorp hybrid fowls, was effective. As with other manifestations of the donor versus host reaction in avian embryos[2,5], the leucocyte fraction of the blood was found to be responsible for this phenomenon; red blood cells and serum were incapable of producing characteristic nodular membrane lesions. It was also observed that the ability of the leucocytes to induce the lesions was abolished by freezing and thawing, and that white blood cells and spleen cell suspensions from embryonic donors were ineffective. Adult fowl spleen and thymus cell suspensions were capable of inducing characteristic chorioallantoic lesions, although thymic cells appeared less effective than spleen cells and leucocytes.

The number of nodular chorioallantoic lesions which developed depended upon the number of cells inoculated on to the membrane. An inoculum containing 10^4–10^5 peripheral blood leucocytes was required to produce a single nodule. When graded concentrations of cell inocula were given, an approximately proportional relationship was observed between the number of cells introduced and the number of nodular lesions produced. Several experiments have been performed with highly in-bred strains of White Leghorn fowls and

embryos. Embryos of the in-bred line responded with approximately the same number of membrane lesions as hybrid White Leghorn-Australorp embryos when inoculated with leucocytes from an unrelated donor fowl. However, when cells from an in-bred donor were placed on the chorioallantoic membranes of isologous embryos, the number of lesions which occurred was far less than the number produced by the same cell suspension on unrelated hybrid embryos.

The investigations made to date indicate that the nodular chorioallantoic membrane lesions result from an immunological reaction between the adult cells and embryonic recipient, presumably of the 'donor versus host' type. The data suggest that the production of lesions requires a significant antigenic disparity between donor and host. In regard to the number of nodules produced, it is a reasonable assumption that each lesion arises as a result of the effective implantation of a single donor cell, but further work will be needed to establish what factors are responsible for the ratio of 10^4–10^5 : 1 between cells inoculated and nodules observed. It may be that a large fraction of the leucocytes are potentially capable of nodule formation but that essentially accidental factors allow only a minute proportion actually to initiate production. At the other extreme, it may be that a very small proportion of cells predetermined to react with the effective isoantigens are responsible for the lesions.

Irrespective of the way the nodules are initiated, it seems legitimate to regard them as arising by proliferation of immunologically competent donor cells in response to antigenic stimulation by components of the embryo. If this view is correct, the phenomenon of chorioallantoic nodule formation should provide a useful approach to certain of the problems of current immunological theory, and may be particularly relevant to the clonal selection hypothesis of Burnet[6].

I wish to express my sincere gratitude to Sir Macfarlane Burnet for his stimulating interest and advice in this work.

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Influenza A′ Infection of Cultured Cells of Whole Mouse Embryos

Cells of whole mouse embryo grown in culture have been used for work with polyoma virus[1,2], and influenza viruses have been grown in cultures of other mammalian tissues such as monkey kidney[3,4], human embryo kidney and lung[5] and bovine embryo kidney[6].

For a recent study of the 'eclipse' phase in the multiplication of influenza *A′* virus, the virus was grown in cells of mouse embryo lung[7]. A greater quantity of tissue can be obtained for culture more easily by using whole mouse embryos, and as is described below, such cultures support the growth of this virus.

Whole mouse embryos taken from stock albino mothers bought from a dealer were weighed, chopped up and treated with trypsin. The cells so released were suspended in a medium consisting of inactivated normal rabbit serum, 1 part, 2·5 per cent lactalbumin hydrolysate, 1 part, and Hanks's balanced salt solution, 8 parts, together with sufficient penicillin, streptomycin and mycostatin to control possible contaminants. Such a cell suspension, containing 5–10 × 10^5 cells per ml., was put into insulin vials in 2 ml. lots and produced monolayer cultures suitable for infection with virus after incubation for 18 hr. at 36·5° C.

Infection was achieved by removing the medium from the cells and, after washing them with balanced salt solution, replacing it with the same volume of a 1 : 50 dilution in 5 per cent normal rabbit serum in balanced salt solution of homogenized mouse lung suspension, representing a later passage of the strain of influenza *A′* used in the earlier work[7]. The cells were held at 36·5° C. for 45 min.; then the infecting suspension was removed and kept for infectivity titration in hens' eggs. The cells were washed in 3 changes of balanced salt solution and their original medium restored. This medium was sampled for infectivity titration 2 hr. and 24 hr. after the first contact of the cells with the infecting suspension.

Table 1. Growth of Influenza *A′* Virus in Cultured Cells of Whole Mouse Embryos, showing Embryo Weights

Experiment	Embryos, Mean wt. (gm.)	50 per cent Egg Infective Dose (ml.) Infecting suspension	Culture medium 2 hr.	Culture medium 24 hr.
1	0·08	$10^{-6.3}$	$10^{-4.7}$	$10^{-3.3}$
2	0·11	$10^{-6.7}$	$10^{-3.5}$	$10^{-3.0}$
3	0·31	$10^{-6.6}$	$10^{-3.3}$	$10^{-4.6}$
4	not recorded	$10^{-6.9}$	$10^{-3.8}$	$10^{-3.6}$

As can be seen from Table 1 the system supported multiplication of the virus, the increase in infectivity for eggs in 22 hr. ranging from 0·5 to 2·8 $\log_{10}$ units. This increase may be compared with the finding that medium removed from infected cells at 2 hr., when its mean 50 per cent egg infective dose in three further experiments was $10^{-3.1}$ ml., was rendered completely non-infective by 22 hr. incubation at 36·5° C. in the absence of cells.

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Action of Antibodies and Plasmin on Ehrlich Ascites Tumour Cells

The mechanism of cell destruction in allergic inflammation is not well understood. Two explanations for production of cell damage have, however, been proposed: (*a*) that the antigen-antibody reaction causes, in some non-specific manner, permeability changes in cells which result in the release of active

mediators of inflammation (histamine, etc.) and that the latter are directly involved in the allergic process[1]; (*b*) that the antigen-antibody reaction activates proteolytic enzymes (plasmin, proteases) that injure the cells[2–4].

The second hypothesis has been supported by some experimental evidence; but the exact mechanism of cell destruction by proteolytic enzymes remains to be elucidated. Studies on the effect of cytotoxic antibodies against Ehrlich ascites tumour cells have revealed that the cytopathogenic changes induced in tumour cells by the action of specific antibodies were very similar to those obtained by the action of streptococcal hæmolysins[5]. It was also found that tumour cells treated with streptococcal hæmolysins were completely disintegrated by the presence of small amounts of proteolytic enzymes[5]. Therefore the action of plasmin was tried on tumour cells affected by specific antibodies, so as to obtain some insight into the role of blood proteolytic enzymes in this process. Rabbits were immunized with homogenates of Ehrlich tumour cells according to the method of Salk and Laurent[6]. Four weeks following the first injection, the sera of these rabbits up to a titre of 1 : 20 caused a distinct cytopathogenic effect on tumour cells. The action of antiserum was found to be dependent on complement.

In a typical experiment 0·1 ml. of washed ascites cells (10^8/ml.) were incubated at 37° C. with 0·1 ml. of immune serum and with 0·8 ml. of *M*/15 phosphate saline buffer *p*H 7·4. Five minutes following addition of antiserum, agglutination of the tumour cells occurred; while 10 min. later first signs of cellular damage could already be seen. The cells appeared swollen, the nucleus became visible and was surrounded by a clear cytoplasmic space with granules close to the nucleus. Thirty minutes following the addition of antiserum, the majority of the tumour cells appeared dead, as established by staining with trypan blue. Sixty minutes following the addition of antiserum the total number of the tumour cells was reduced by 25 per cent (Table 1). The remaining cells, though swollen, retained their cytoplasmic membrane, and the nucleus appeared undamaged. Only a very small amount of cell debris could be seen. These results are consistent with the findings of Flax[7] and Goldberg and Green[8], who tested the action of antiserum against Ehrlich and Krebs ascites tumours respectively. The action of human and rabbit plasmin, activated by streptokinase *SK*–100 units/ml.

Table 1. Action of Antiserum and Human Plasmin on Ehrlich Tumour Cells

Reaction mixture	Cells remaining* (per cent)
Tumour cells in buffer[a]	100
" " + 10 per cent antiserum[b]	75
" " + 10 per cent inactivated antiserum[c]	100
" " + Plasmin (250 U/ml.)	100
" " + 10 per cent antiserum + plasmin (1 U./ml.)[d]	54
" " + 10 per cent antiserum + plasmin (10 U./ml.)	10
" " + 10 per cent antiserum + plasmin (20 U./ml.)	1
" " + 10 per cent antiserum + plasmin (50 U./ml.)	0·01
" " + 10 per cent antiserum (inactivated) + plasmin (50 U./ml.)	100
" " + 10 per cent antiserum + streptodornase (1,000 U./ml.)	100

* Average of the counts of 3 experiments after 60 min. incubation at 37° C.
All the cells that still possessed a cell membrane, no matter how damaged they appeared, were counted.
Usually 500 cells were counted in each experiment.
[a] Ehrlich tumour cells 10^7/ml. washed with phosphate-buffer in saline, *p*H 7·4.
[b] Rabbit anti-Ehrlich serum.
[c] Anti-Ehrlich serum heated to 56° C. for 30 min.
[d] Tumour cells were incubated with antiserum for 15 min. at 37° C. before addition of human plasmin. Similar results were obtained with plasmin prepared from rabbit's serum.

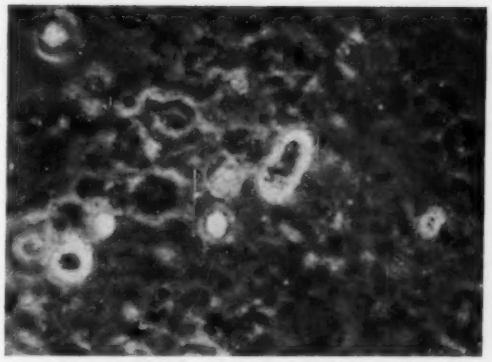

Fig. 1. Ehrlich ascites tumour cells incubated for 15 min. at 37° C. with antiserum and plasmin (50 U./ml.). Plasmin was added to tumour cells after they were in contact for 15 min. with antiserum. Note the massive destruction, accumulation of cell debris, few tumour cell ghosts and several undamaged leucocytes. (× 450)

(Varidase-Lederle), was tested on tumour cells affected by antiserum. Rabbit and human plasminogen were prepared according to the method of Ratnoff[9]. In addition, antiserum was also used as a source of plasminogen. Plasmin was assayed according to the method described by Remmert and Cohen[10]. Plasmin (1–50 units/ml.) was added to Ehrlich tumour cells which had been pre-incubated for 15 min. at 37° C. with antiserum, and which showed distinct swelling. As soon as 10 min. after addition of plasmin, the swollen cells began to disintegrate, and 5 min. later large accumulations of cell debris mingled with ghosts of swollen cells could be seen (Fig. 1). Incubation with increasing concentrations of plasmin caused a proportional increase in the percentage disintegration of the tumour cells (Table 1). Under the same experimental conditions neither plasmin alone (250 units/ml.) nor plasmin incubated with heat-inactivated antiserum (56° C. for 30 min.) or streptodornase (1,000 units/ml. present in 'Varidase' preparations) had any effect on the morphology or total number of cells (Table 1). The leucocytes which were present in the tumour suspensions incubated with antiserum and plasmin appeared undamaged (Fig. 1). If on the other hand plasmin was added to tumour cells either simultaneously or 5 min. after addition of antiserum (before any apparent damage to cells occurred) only few cells appeared swollen and no disintegration of tumour cells was observed. These results are consistent with the findings of various investigators[11] that plasmin destroys complement. This assumption seems plausible since the action of cytotoxic antibodies against Ehrlich tumour cells have been found to be dependent on complement[7]. Thus, to obtain rapid disintegration of cells by the combined action of antiserum and plasmin the antibody-complement complex must first damage the cell membrane. This leads to altered permeability of the cell membrane which may allow plasmin either to penetrate into the cells or to attack cell wall substrates which have been unmasked by the action of antiserum. This would result in the depolymerization of cell components leading to cell disintegration.

The action of plasmin was found to be unspecific as both trypsin and papain were active in this respect. These *in vitro* findings raise the possibility that plasmin or other tissue proteolytic enzymes may be directly involved in cell destruction induced by the antigen-antibody reaction.

Work is in progress to determine what part, if any, these factors play in cellular damage *in vivo*.

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Dietary Fats and Thrombosis

In order to study the influence of the fat content of food on the occurrence of thrombosis in the arterial and venous system, a group of 133 hospital patients aged 65–90 years were treated with a diet in which butterfat, margarine and lard were replaced by vegetable oils (unhydrogenated corn oil and soybean oil). The daily intake of vegetable oils was about 40 gm., or half the total fats. The time of treatment of the 133 patients ranged from 1 to 17 months (average 4·7 months) representing a total of 52½ years. For comparison, a control group of the same size and composition who received an ordinary hospital diet containing about 80 gm. animal fats (including about 40 gm. butter-fat, margarine and lard) was used. The occurrence of thrombosis and embolism in both groups was diagnosed clinically and post-mortem.

In the treated group only 4 cases of thrombo-embolism were observed: 1 case of myocardial infarction after 6 days of treatment, 1 case of myocardial infarction after 26 days of treatment, both verified by autopsy; 1 case of little stroke with transient aphasia of a few days' duration occurring after 30 days of treatment, and 1 case of pulmonary embolism after 4 months of treatment in a patient with bronchial cancer (verified by autopsy).

In the control group 15 cases of thrombosis were observed: 2 cases of thrombophlebitis (1 case complicated with pulmonary embolism and 1 case with thrombosis in the carotid artery), 2 cases of pulmonary embolism (both verified by autopsy), 5 cases of myocardial infarction (all typical, 2 verified by autopsy), 6 cases of cerebral thrombosis (3 verified by autopsy).

The difference in the occurrence of thrombosis in the two patient groups was statistically significant ($t = 5{\cdot}1$) and cannot be interpreted in terms of variations in age, weight and blood pressure in the groups.

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BIOLOGY

Interaction between the Plasma of the Loggerhead Turtle and Toxin of the Portuguese Man-of-War

The loggerhead turtle, *Caretta caretta*, is reported to eat the Portuguese man-of-war, *Physalia physalis*[1–3]. Where *Physalia* are assembled in windrows, loggerheads are often seen at the surface with eyes closed, eating this siphonophore. *Velella*, an allied species, has also been identified from stomach contents of a turtle which was identified as a loggerhead[4]. *Physalia* is equipped with a singularly potent toxin[5]. This is contained in nematocysts which can penetrate a rubber glove and presumably even the chitinous exoskeleton of a crab's leg. As Barbour pointed out, the dense impenetrable skin of the face and jaws might protect a turtle from some of the obvious hazards of such a feast. However, since tentacles of active colonies have been observed to cling tightly to the skin, gloves, glass containers and even to the polished steel surface of a pair of scissors, it is difficult to explain the ability of the turtle to swallow, much less to digest and survive this meal. We believe that the acid of the stomach contents would inactivate the nematocyst and its contained venom virtually upon contact. The unconcern of the adult turtle with the toxic effects of the nematocysts might be due to the presence of protective antibodies in the blood acquired by contact with *Physalia* early in life. It is the object of this communication to report the results of experiments designed to test this hypothesis.

A 15-kgm. loggerhead turtle which had been in captivity less than one week was obtained from the Miami Seaquarium. The jugular vein was cannulated and the animal was bled. Sufficient potassium oxalate was added to the blood to bring the concentration to 0·2 per cent and the blood was cooled. Later the blood was centrifuged and the separated plasma was frozen at −20° C. without preservative. Toxin of *Physalia* was prepared according to procedures already described[5], with the following modifications: (1) Washed quartz sand and glass-distilled water were used for homogenization of the washed purified nematocysts. (2) After centrifugation, the material was mixed with 4/3 volumes of calcium phosphate gel prepared by the method of Kunitz[6] and was recentrifuged. The water-clear supernatant solution apparently contains all the toxic material. (3) This supernatant solution was lyophilized in tared 5-ml. capsules. These were sealed without admitting air and stored at −20° C. until used.

Fractions prepared in this way have regularly assayed at *LD*90 of 2·5–3·0 mgm./kgm. body-weight of pure strain, 18–25 gm. *C*57 black male mice when the mice are injected intravenously. The lyophilized material dissolves easily and completely. We assume that the entire weight of lyophilized material is toxic for purposes of estimating concentrations. It should be noted that if any significant fraction of the lyophilized 'toxin' were inactive then the active moiety would considerably exceed 2·5 mgm./kgm. in toxicity. The toxin is quite thermolabile and its activity decreases during manipulation.

In one experiment lyophilized toxin was dissolved in glass-distilled water to produce a concentration of 20 mgm./ml. One aliquot was diluted with 19 vol. of 0·9 per cent saline to a final concentration of 1 mgm./ml. (solution *A*, Table 1). The second aliquot was diluted with one volume of turtle plasma

Table 1

Solution	Toxin mgm. (dry weight)	Total diluent added: Plasma (ml.)	Water (ml.)	0·9% Saline (ml.)	Final toxin concentration (mgm./ml.)
A	20	0	1	19	1
B	20	1	1	18	1
C	20*	1	1	18	1*
D	20*	1	1	0	10*

* Weight of toxin expected if it were all present in the supernatant solution (*C*) or in the precipitate (*D*).

and then further with 18 vol. of saline to a final toxin concentration of 1 mgm./ml. (solution *B*, Table 1). A precipitate formed when the plasma was combined with the toxin which did not dissolve when more saline was added. This precipitate was uniformly dispersed through the injection fluid and was included in the assay. The third aliquot was combined with an equal volume of turtle plasma and centrifuged to remove the precipitate. The supernatant liquid was assumed to contain all the toxin and was so diluted with saline as to bring the final toxin concentration to 1 mgm./ml. (solution *C*, Table 1). The precipitate was resuspended in a solution consisting of 1 ml. of turtle plasma and 1 ml. of glass-distilled water (solution *D*, Table 1). If all the toxic material were contained in the precipitate the concentration would have been 10 mgm./ml. Table 2 presents the results of assays of these solutions.

Table 2

Solution	No. mice injected	Dose-range (mgm./kgm.)	Surviving after 2 hr.
A	10	2·55–2·61	0
B	10	2·56–2·59	0
C	11	2·57–2·61*	0
D	10	47·0 –56·2*	10

* Weight of toxin expected if it were all present in the supernatant solution (*C*) or in the precipitate (*D*).

Apparently all the toxicity originally present in solution *A* is still present in the toxin-plasma combination *B*. The precipitate which was separated and resuspended in saline *D* contained none of the activity. The supernatant solution *C* held all the toxicity of the original combination.

This experiment was repeated twice, with fewer mice, with entirely similar results. These results suggest that the loggerhead turtle lacks blood immune bodies which might be invoked to explain its apparent insensitivity to the toxin of *Physalia*.

Our interpretation does not exclude the possibility that the loggerhead might possess localized tissue antibodies. Phisalix[7] has shown that reptiles, and especially turtles, are less sensitive to the sting of bees than other vertebrates. It is possible that turtles are not susceptible to venoms of the type injected by *Physalia*.

We are grateful to Dr. Wilhelmina Dunning of the University of Miami for providing mice and to the Miami Seaquarium for the loggerhead turtle. These studies were supported by Grant *RG*-5272 from the U.S. Public Health Service, National Institutes of Health.

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Elucidation of the Life-cycle of *Fasciola hepatica*

Dr. B. Dawes, in his recent communication[1], has made an important contribution to our knowledge of the trematodes. In the interests of historical accuracy and in justice to a great nineteenth-century zoologist I would, however, like to point out that the life-histories of several trematodes, including that of *Fasciola hepatica*, were beautifully and accurately described by J. J. S. Steenstrup in his work "On the Alternation of Generations", translated into English and published as a Ray Society monograph in 1845, nearly forty years before the work of R. Leuckart and A. P. Thomas. Steenstrup solved one of the most difficult of all zoological problems, and his ideas on the alternation of generations were considered so revolutionary that G. Buck, his translator and editor, thought fit to write an apologetic introduction to assuage the anger of his readers.

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I agree with H. S. Jefferies that historical accuracy is desirable and that the credit for scientific discovery should go where it is deserved. Parasitology owes much to the work of Steenstrup, who demonstrated the 'alternation of generations' which occurs in the lives of some invertebrate animals[1]. This expression was first used in 1819, however, by the Franco-German poet and naturalist, Louis Adelaide de Chamisso, in regard to the life-cycle of the pelagic tunicates known as salps, but the young Danish zoologists showed that it applies to the life-cycles of some cœlenterates, trematodes and other invertebrate animals as well. Valuable discoveries were made also by other zoologists, and some of these were mentioned in Chapter 16 and elsewhere in my book[2], and others have since been indicated by E. G. Reinhard, with special reference to the life-cycle of *Fasciola hepatica*[3].

In using the term 'cercaria', Steenstrup recognized both a discovery and an invention made in 1773 by O. F. Muller. The forms which he called 'nurses' were later named 'rediae' by F. de Filippi in honour of Francesco Redi who, in 1668, made all discoveries in this field possible by destroying the false doctrine of spontaneous generation. Such 'nurses' were first seen in 1818 by Ludwig Bojanus, who called them 'royal-yellow worms' and emergent cercariæ were first seen to encyst in 1807 by C. L. Nitzsch of Halle, who thus witnessed the formation of what Steenstrup later called 'pupæ'. Some earlier discoveries made by two of C. A. Rudolphi's pupils did not enter into Steenstrup's considerations, though they concern the trematode life-cycle. In 1831, K. E. Mehlis described the hatching of 'ciliated embryos' from the eggs of trematodes, and in 1837, F. C. H. Creplin observed that such forms (miracidia) emerge from the eggs of *Fasciola hepatica*. Unluckily, Bojanus, Steenstrup and other persons mentioned here by name seem not to have made a study of the parasites of *Limnæa truncatula*, and for this reason did not observe the larval stages of *F. hepatica*. Steenstrup made a brief reference (5 lines) to the adult of this species and then turned to descriptions of echinostome and stylet cercariæ. He did not even mention the larval stages of the liver-fluke, and in fact he did

not describe any trematode life-cycle in its entirety. According to Reinhard, the German parasitologist, Davis F. Weinland was the first person to suspect that the larval stages of the liver-fluke occur in *L. truncatula*. In 1875, he "told of finding cercaria-sacs" in the digestive gland of this snail, and he also noted that the cercariæ tended to leave water, which prompted him to suggest that they encysted on herbage and were the young of *Fasciola hepatica*. Thomas was apparently unaware of Weinland's discovery and conjectures, and his well-deserved success was the result of a methodical search for possible snail hosts in localities where sheep were liable to contract liver-rot. The quest was probably simplified by the knowledge that only fishes and molluscs were to be regarded with suspicion because the only animals known to harbour larval trematodes, but success called for all the resources of a most resourceful zoologist.

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Differentiation induced as Season Advances in the Embryo-Gametophyte Complex of *Pinus nigra* var. *austriaca*, using Indole Acetic Acid

Maintenance of growth in very young excised gymnosperm embryos in culture medium has been attempted by one of us with limited success. Indefinite growth was never achieved, and at death, differentiation was lacking other than that suggesting initiation of symmetry or polarity.

The present work on *Pinus austriaca* arises out of attempts to find qualitative evidence of physiological relationship in the region of the parental-embryonic threshold. Indole acetic acid, because of its acceptance as a growth-controlling agent, was added to the culture medium in the hope that it would stimulate differentiation during post-zygotic *in vitro* life.

Seeds were gathered at intervals during the year. Their testas were removed under aseptic conditions, and the naked gametophytes were placed on a sterile culture medium buffered to *p*H 6·0, and containing a known concentration of indole acetic acid. Various concentrations of indole acetic acid were sampled in the course of the experiments.

The gametophytes were harvested, some after two, others after four weeks *in vitro*.

Examination followed sectioning and staining in borax carmine and fast green.

Comparison of several gametophytes grown on an indole acetic acid medium in July immediately following fertilization showed in each case the presence of a meristematic layer crossing the gametophytic tissue. Position of the meristem varied from one example to another. It consisted of contiguous thin-walled cells resembling the fusiform initials of a vascular cambium.

Cells between the periphery of the gametophyte and the meristematic layer had undergone enlargement which produced local swelling. These cells contained little cytoplasm and only a few granules. Cells on the other side of the layer were much smaller, irregularly shaped, and contained a strikingly large number of granules which differed markedly in form from those normally present at corresponding positions in controls.

In many of these gametophytes, the parenchyma cells surrounding the archegonial jackets developed secondary thickenings on all their walls. Thus in these cells pits appeared on all the walls; they were large and circular.

Experiments performed in August, long after fertilization, showed neither the development of meristematic layers in gametophytic tissue nor the development of thickenings and pitting outside the archegonial jackets. Therefore, none of the phenomena arising in July examples occurred in the August ones.

In the older examples a different feature arose not seen in the July ones. At several scattered locations, cells one to three times the volume of neighbouring ones arose. These were thick-walled, lignified, pitted and often anisotropic in polarized light. Cytoplasm and nuclei were not visible in them.

Proembryo ontogeny for indole acetic acid *in vitro* cases differed from that for the controls in both the July and August examples. One cultured proembryo developed reticulate pitting in the walls of the suspensor cells. Others showed inhibited growth. Generally, on the basis that incidence of anomaly and differentiation was less frequent for embryo than for gametophytic tissue, the effect of the indole acetic acid *in vitro* conditions was judged to be less marked for the embryo system.

Experiments performed at the end of March with material collected at that time (nine months after fertilization) showed none of the previously described phenomena.

It may be concluded that: (1) In *Pinus austriaca*, as one example of an embryophyte, parental-embryonic environment shows a differential response to a factor stimulating differentiation. This differential suggests presence of a pattern of organization in which time and morphological events are significant and related. (2) In parental tissue, visual differentiation may be extensive; for example, meristem and massive activity near archegonia, or confined to single isolated cells, for example, lignified structures. In embryonic tissue the effect is always confined to single cells. (3) Indole acetic acid appears to be influential in initiating the differentiation and in effecting its course.

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Collection of Urine and Fæces from the Chicken

In order to examine some factors which influence the intestinal absorption and urinary excretion of calcium and phosphorus in the chicken, a surgical method for the separation of urine and fæces has been developed which depends upon the successful 'exteriorization' of the distal end of the large intestine. This method, which has been employed principally in adult Rhode Island Red females, is equally suitable for adult capons and cockerels. Females operated on one year ago and fed practical

rations have maintained normal behaviour, appetite, laying performance and live weight compared with non-operated controls. They have been employed for the daily collection of urine and fæces in various experiments, to be reported elsewhere, ranging from three to ten weeks duration.

Exteriorization of the distal end of the large intestine at a point on the abdominal wall has been previously reported by others. Rothschild[1] reviewed previous reports of techniques while contributing his own method, but mentioned no criteria of health and production in operated birds. Ariyoshi and Morimoto[2,3] described a technique that was afterwards used for nitrogen balance studies in cockerels over a period of six months and which necessitated the use of a special diet. Theirs is the only report of a long-term metabolic study on separated urine and fæces in the chicken.

The principal criticisms, therefore, of the previously described methods are: (1) they fail to provide such criteria of normality as behaviour, appetite and production; (2) there has been insufficient emphasis on, or attempt to explain, the post-operative complications and how to avoid them; (3) none has mentioned the quality of the fæces, which are invariably scant and fluid if there is any degree of stenosis or intestinal atony; (4) no long-term quantitative observations with laying females have been reported.

In carrying out the operation, general anæsthesia and full aseptic methods are essential. The terminal end of the large intestine as it joins the cloaca is ligated, incised, and exteriorized at a point half-way between the ischium and the abdominal mid-line approximately one and a half inches anterior to the ventral lip of the cloaca on the right side. The cloaca now forms a blind sac (except for the oviducal orifice in the female), and urine, which enters it from the ureters, is periodically expelled since the normal defæcation reflexes are retained. To collect the urine, a wooden frame covered with polythene sheet is placed below the wire floor of the cage. The polythene sheet slopes towards a central hole through which the urine flows into a vessel. The fæces, which are firm in consistency and in short lengths, pass out of the exteriorized bowel continually. To collect the fæces, a 100-ml. polythene beaker is inserted into a rubber cuff with a roll end sutured to the skin around the exteriorized bowel.

The production of a successful preparation depends upon the elimination of several post-operative complications. Any degree of stenosis due to inflammation, fibrosis, contraction of abdominal muscle or fascia and skin overgrowth around the exteriorized bowel must be avoided. Stenosis due to any of these factors always induces fæcal obstruction, constipation and intestinal atony. In order to overcome these complications, a thin-walled glass cannula, with approximately the same diameter, is inserted into the exteriorized bowel immediately after the operation. This cannula, through which fæces pass freely, is held in position by a polythene collar sutured to the skin immediately surrounding the exteriorized bowel. After two weeks, by which time contraction around the exteriorized bowl has terminated, the cannula is removed and fæces continue to pass quite freely. In some birds the skin immediately surrounding the bowel tends to overgrow the bowel orifice, but this difficulty is overcome by judiciously cutting it away. The birds remain clean and require little attention except in the course of experiments, when it is necessary to ensure that the fæces collector is securely attached.

I wish to express my thanks to members of the surgical unit at the Cambridge University School of Veterinary Medicine for their suggestions and criticisms, and I am indebted to the British Oil and Cake Mills, Ltd., for the provision of a research studentship which enabled this work to be carried out.

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Infection of Peas by Wilt Disease Fungi

A serious soil-borne wilt disease of peas has been shown to be caused by *Fusarium oxysporum* f. *pisi* (Linf.) Synder and Hansen, and *Fusarium oxysporum* Fr. emend. Synder and Hansen var. *redolens* (Wollenw.) Gordon[1,2]. Both these fungi attack the vascular system of the plant, and a typical internal symptom of the disease is an orange or brick-red discoloration of the vascular tissue.

During the examination of wilted pea plants which had been grown from seed in naturally infested soil it was noticed that, when the lowermost leaves began to wilt, which is the earliest recognizable stage, no discoloration could be found except in the cotyledonary vascular bundles. Furthermore, in plants which showed the curious 'half-leaf' wilt in which half of each leaf was wilted, only one of the two cotyledonary bundles was orange. Following these observations a series of isolations were made from the steles of plants showing various degrees of wilting. All plants with any sign of foot-rot (*Fusarium solani* f. *pisi*) were discarded, and as this fungus was not isolated from any of the experimental plants it is considered that the results in Table 1 refer to the wilt disease alone.

Table 1. Distribution of the Fungus in the Plants at Various Stages of Wilting

Leaves		Stem	Cotyledonary bundles		Hypocotyl	Root
Total	Wilted					
8	0	−	−	−	−	−
8	1	−	+	+	−	−
8	2	−	+	+	+	−
8	4	−	+	+	+	+
8	5	+	+	+	+	+
8	6	+	+	+	+	+
Completely wilted plant		+	+	+	+	+
'Half-leaf' wilt		+	+	−	+	+

An examination of Table 1 reveals that in the earliest recognizable stages of the wilt disease the fungus can be isolated from the cotyledonary bundles only; but as the wilt symptoms progressed so did the distribution of the fungus. Where the early stage of 'half-leaf' wilting was present the fungus was isolated from one cotyledonary bundle only. Consideration of the configuration of the vascular system of the pea plant[3] indicates that from a unilateral infection this 'half-leaf' wilting might be anticipated.

Microscopical examination of the distribution of the fungus within the plant confirms the foregoing

observations. In 'half-leaf' wilt the fungus extends much farther up the vessels of the wilted side of the stem than of the non-wilted side, and in the root of an extensively wilted plant only two of the three points of the triarch stele, those corresponding to the cotyledonary bundles, contained the fungus.

It was concluded, therefore, that, in peas grown from seed in naturally infested soil, invasion of the host by the fungus is through the moribund cotyledons. Occasionally, where the root has had to curl around the seed before growing downwards, infection has been observed at the junction of an upper lateral root with the tap root. In plants in the first stages of wilting, infection at other points has been recorded in a very few cases, but it has always been associated with other damage, for example, by eelworms.

Field observations confirm these findings but, on land which has been too frequently cropped with peas, the root disease picture is often complicated owing to the multiplication of other soil-borne pests and diseases.

This investigation is continuing, and a more detailed report will be published later. I am indebted to the Agricultural Research Council for a grant enabling me to do this work, to Sir Joseph Hutchinson for laboratory and glasshouse facilities and to the National Agricultural Advisory Service for their co-operation.

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Arm Abnormality in *Octopus*

While working through the cephalopod collections of the University of Miami Marine Laboratory I found a specimen which possessed an abnormal third left arm. The arm was divided into two branches. An unusual feature was the presence of a well-developed web between the two divisions as found uniting the bases of normal arms in this species.

Regeneration in cephalopods has been observed and reported by several workers but they are generally concerned with unipedal growth as a result of damage or autonomy. A. E. Verrill[1] reported on the regeneration of suckers in the giant squid, *Architeuthis harveyi*. Parona[2] described the occurrence of two, thin, short arms in the position of the first left arm of *Octopus vulgaris*; the presence of an extra arm between the second and third right arm in *Eledone aldrovandi*; and the vague mention of a divided arm in a specimen of *Eledone moschata*. Hanko[3] cited the occurrence of a split right fourth arm in a specimen of *Octopus vulgaris*. M. Lange[4] concerned herself with the histological processes and method of regeneration and only cited Hanko's 1913 work without further comment. Robson[5] mentioned the occurrence of regeneration, Lange's work, and autophagy. Sasaki[6] noted and figured a monstrosity of *Octopus vulgaris* in which the right fourth arm gave rise to 25 offshoots, all bearing suckers but with no indication of a web. He stated that he had observed such abnormality in two cases, both *Octopus vulgaris*. S. G. A. Jaeckel[7] reproduced Sasaki's figure.

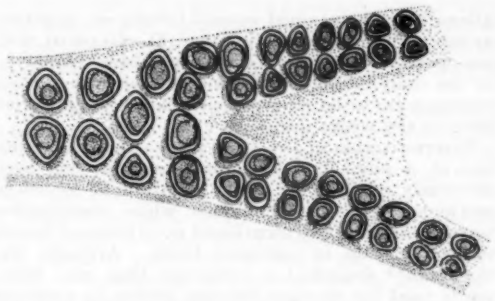

Fig. 1. Ventral view of the split-arm specimen of *Octopus briareus* Robson

The specimen (*Octopus briareus* Robson, 1929, U.M.M.L. 31 : 160) examined by me appears unique in the presence and extent of a web between the two extremities of the split third left arm. An existing web between extra arms has not been recorded previously or at least has escaped notice of the above-cited workers.

The specimen examined was a male, collected by Clench and Dickinson on June 7, 1956, at Big Pine Key, Florida. It was identified by means of the key and description given by Voss[8] for *Octopus briareus*. Basic measurements and indices are given in Table 1. Indices used are as defined by Pickford[9] and Voss[8].

Table 1. Measurements (mm.) and Indices of the Split-arm Specimen of *Octopus briareus* U.M.M.L. 31 : 160 (Male)

Mantle length	51·2
Mantle width	39·5
Mantle width index	77·1
Hectocotylized arm length	173·3
Ligula length	5·6
Ligula index	3·2
Arm order	3.2.4.1

The length of the third left arm (left ventrolateral arm) as measured from the first basal sucker to the junction of the split is 94·3 mm. When the arm is viewed ventrally (sucker side up) the split sections measure 101·5 mm. and 103·5 mm., left and right side respectively.

The suckers are biserial in the normal fashion from the mouth to immediately below the junction. The second row of suckers, distally from the centre of the junction, is composed of three suckers across. The last row of suckers on the main stem has four suckers across the arm. On each of the divided sections the paired sucker arrangement found on the main stem is present. The web depth is 8·2 mm. between the two branches. A ventral view of the split arm is shown in Fig. 1. Measurements of the split arm are summarized in Table 2.

Table 2. Measurements (mm.) of the Split Third Left Arm

Arm length (to base of split)	94·3
Arm length of left branch	101·5
Arm length of right branch	103·5
Depth of web	12·1
Diameter of largest sucker	7·3

This type of growth does not appear to be the result of an arm injury. The web is shallow in this species and the presence of a web (11 per cent of the branched arm length) between the split sections would seem to indicate a mutant or an abnormality caused by injury to the embryo. No adaptive use for this development can be seen as the ligula is well developed and all other arms are intact, nor did the modification appear to be detrimental to the animal as it was in

good shape and apparently healthy at the time of capture.

I wish to acknowledge the constructive criticism offered by Dr. G. L. Voss.

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Delayed Birth in Marsupial Macropods—the Euro, the Tammar and the Marloo

THE first anecdotal evidence for delayed birth in macropods was given by Carson[1] in 1912. However, the phenomenon of delayed birth in the quokka (*Setonix brachyurus*) and the tammar (*Protemnodon eugenii*) was first investigated in detail by Sharman[2-4].

During a study of the euro (*Macropus robustus*) undertaken by the University of Western Australia we have found many delayed blastocysts lying free in the uteri while the females had 'joeys' in the pouch. The blastocysts were located by flushing out the uteri with physiological saline as described by Sharman[3]. Following this discovery an attempt was made to initiate a delayed birth in an experimental manner. A female euro obtained from the local zoological gardens was found to have a 28-gm. 'joey' in the pouch. This 'joey' was removed and the female isolated from further contact with males. 36 days later examination of this animal revealed a pouch young attached to the teat. Examination on the thirty-fourth day had shown no pouch young.

Previous work by Sharman[4] showed that tammars contain delayed blastocysts, and he has given anecdotal evidence which supported a delayed birth mechanism in this species. We have also produced a delayed birth pouch young in the tammar by removing a 0·70-gm. 'joey' and segregating the female from all males. Unfortunately, initial examination of the pouch was made 62 days after the removal of the first 'joey'.

During the course of the present work on marsupial macropods we have found blastocysts lying free in the uteri of two marloos (*Macropus rufus*) which both had 150-gm. 'joeys'. Apart from Carson's[1] initial observation this appears to be the only reported evidence for delayed birth in this species.

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ENTOMOLOGY

Time of Feeding in Relation to the Toxicity of a Stomach Poison to the Desert Locust

PEARSON[1] first showed that the stomach toxicity of dieldrin to fifth-instar hoppers (nymphs) of the desert locust (*Schistocerca gregaria* Forsk.) was increased by a period of starvation after treatment and suggested that this was due to variation in the rate of passage of the poison through the gut. We have confirmed and extended his observations and are directing attention to them because there appears to be little published information on the effects of starvation on the toxicity of stomach poisons to insects[2] although attention has been given to the effects of the diet fed to insects before they are used for experimental purposes[3].

When insects are fed with poisons applied by methods (for example, the poisoned sandwich method) other than those utilizing a poisoned medium there are usually time intervals between removal of the insects from their normal food, consumption of the poison, and resumption of normal feeding. In practice the insects are usually starved before an experiment, in order that they will readily accept proffered food and/or poison[2]. When the time-intervals are variable and of the duration of several hours they can affect the rate of passage through the gut of small quantities of food such as those that comprise a 'sandwich' containing poison.

A batch of fifth-instar nymphs, 4–7 days after moulting, was randomly divided into three groups. Two of the groups were deprived of food for 24 hr. Each insect in all three groups was then fed a small quantity of fluorescent powder ('phosphorescent powder, green', supplied by Rex Campbell, Ltd.) applied in gum to a small piece of grass. Immediately after eating this material two of the groups, including one of those previously starved, were then supplied with abundant fresh grass. The three groups of insects therefore received different treatments: group I was starved before dosing and fed after, group II was fed before and after, while group III was starved before and after. The fæcal pellets produced by each group in successive hours after ingestion of the indicator substance were counted. Each pellet was then broken up in about 1 ml. of water and examined in ultra-violet light, when any granules of the indicator present were clearly visible. The results obtained are shown in Table 1.

Table 1. TOTAL NUMBER OF FÆCAL PELLETS AND NUMBER CONTAINING INDICATOR AT DIFFERENT TIMES AFTER DOSING (PER GROUP OF 10 HOPPERS)

Treatment group	Hours after dosing				
	0–1	1–2	2–3	3–4	5–6
I	7/19	23/31	14/30	18/32	5/36
II	*	2/22	8/18	15/16	16/30
III	0/3	0/3	0/4	3/3	7/7

* Owing to difficulty in making these insects feed, no estimate can be given for this period.

It will be seen from Table 1 that the proportion of pellets containing the indicator depends on the treatment given. However, the quantity of indicator substance in each pellet was very variable, and rough quantitative estimates were made visually of the amount lost per hour. It was estimated that 80

per cent was lost in less than 2 hr. in group I, in 4–5 hr. in group II, and in 7 hr. in group III.

In further experiments the same range of doses of the insecticide dieldrin was fed to each of two groups of hoppers which had been starved for a period of 24 hr. The insecticide was applied to a small portion of a blade of grass, and within 15 min. of eating this the hoppers of one group were supplied with abundant freshly cut grass. No deaths occurred within five days among the 56 insects in this group. The second group was kept without food for a further 24 hr. after dosing and a total of 48 deaths occurred during the next five days from the 55 insects that were dosed. These results corresponded to $LD50$'s of $\geqslant 4{\cdot}8$ and $0{\cdot}79$ μgm./gm. respectively. In the second experiment similar results were obtained, that is, $LD50$'s of $\geqslant 4{\cdot}8$ and $0{\cdot}95$ μgm./gm.

Pearson's toxicity results clearly fit in with these findings. His normal treatment most nearly followed group I above, since his insects "were not fed on the previous evening" but were fed immediately after dosing, while his starvation treatment was equivalent to group III. As a result of the different treatments the resistance of the insects fed immediately after dosing was increased 1·5–2·8 times.

It is interesting to note that what might appear to be a convenient laboratory procedure may yield very variable results. For example, locusts may be deprived of food for 24 hr. before a stomach toxicity experiment is due to be carried out and then given the poisoned food. Later, when convenient, they are supplied with abundant grass ; if this occurs immediately after the poison is consumed, the toxicity will be very much less than if there is a delay of a few hours, as may well occur when a large series of individuals is being treated.

In the field, Ellis and Ashall[4] found that in some cases hoppers exhibited a noticeable diurnal rhythm in feeding, there being least feeding during the 6 hr. after dawn, and most between 1600 and 2200 hr. It is thus probable that when dieldrin is sprayed on vegetation, a technique being used on an increasing scale[5] for locust control, its toxicity will vary with the time of day at which it is eaten.

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Fertilization of Eggs of the Desert Locust by Spermatozoa from Successive Copulations

In the desert locust (*Schistocerca gregaria* Forsk.) copulation occurs frequently, although once fertilization has taken place further copulation appears to serve mainly as a stimulant to egg-laying and to be unnecessary for renewed fertilization[1]. An interesting problem arises as to whether, after a second copulation, eggs are fertilized by sperm from the first or the second copulation. The following experiment was devised, using an albino stock of *Schistocerca*. It has been shown previously that crosses between albino and normal adults show a Mendelian pattern of inheritance, in which albinism is recessive[2].

Four pure albino females were kept in isolation in cages, and when they were sexually mature an albino male was introduced into each cage for 24 hr. and then removed. Subsequent oviposition produced averages of 60–80 young locusts per egg pod, and these included both males and females, showing that copulation had taken place. After each albino female had laid two egg pods, from which only albino hatchlings resulted, a normal-strain male was placed with each of the females and then removed after 24 hr. All subsequent egg pods produced young locusts that were normal in appearance, to the almost complete exclusion of albinos. In fact, out of 1,033 eggs that hatched, 1,028 were normal and five were albino. In a further experiment, the same procedure was adopted except that the albino female was fertilized first by a normal male and then by an albino ; the progeny of the first two pods were all normal in appearance, but after the second copulation all, without exception, were albino.

It is known that spermatozoa can fertilize eggs at least ten weeks after the last copulation[1], but from the present experiments it appears that a second injection in some way prevents further use of the previous spermatozoa. This is evidently not due to competition between a more and a less virile strain, as it occurred whether the albino or the normal male was used second. Whether the first spermatozoa were inactivated in some way, or even killed, by the second, or the second spermatophore mechanically prevented the first spermatozoa from reaching the eggs, cannot be deduced from these observations.

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OCEANOGRAPHY

Manganese Deposition on the Shells of Living Molluscs

Recently, there has been a renewal of interest in manganese deposition on the sea bed, particularly that brought about by biological rather than inorganic processes[1–5]. During the past fifty years[1–14] various theories have been put forward concerning the method of manganese deposition, yet the only measurements on the rate of deposition are those of Petterssen[11,12], and Buttler and Houtermans[13] on manganese nodules of deep-sea origin. These authors give a rate of the order of 1 mm. in 1,000 years.

During a survey of the sublittoral fauna of the Clyde sea area from 1949 onwards, it has been observed that the shells of many living molluscs are encrusted with a black deposit. In particular, this was the case of molluscs taken from muds and sandy-muds at depths of 15–105 fathoms. Although this has been recorded before[7–9,15], past analyses were limited to nodules, and deposits on stones and empty shells. While many specimens of living molluscs in the Clyde have manganese deposited on their shells, it is significant that deposits are more common and much heavier on shells with a dull, matt periostracum than on those with a glossy periostracum.

Yellow, and olive-coloured shells seem to be most affected, although deposits occur on shells of other colour. Thus species of *Nucula* clearly show the difference between glossy and matt surfaces, for while matt *N. sulcata* and *N. nucleus* may be completely covered with the black deposit (none of the older shells of many hundreds examined is free of deposit), glossy *N. turgida* and *N. tenuis* from the same substratum and of the same age rarely show any deposit. When present in the two latter species the deposit is confined to a small, thin film at the umbo. Although the list is not exhaustive, other species noted for their black deposit are *Corbula gibba*, *Venus casina*, *Nuculana minuta*, *Cyprina islandica*, *Mysella bidentata*, *Glycymeris glycymeris*, the underside of *Pecten maximus* and *Chlamys septemradiata*, *Natica alderi*, *Neptunea antiqua* and the tips of *Dentalium entalis*. All these species live at or close to the surface of the bottom sediment.

Analysis confirms that a high percentage of manganese is present in the deposits on these shells, far more than in the bottom sediment in which they live (Table 1). The percentage of manganese is close to that estimated for nodules from the same area. As figures for the rate of growth of *Nucula* species are available[16] micrometer measurements of the thickness of the deposit at the umbo give some idea of the rate of deposition. Shells of *N. sulcata* and *N. nucleus* less than 5 years old have a thin coating that is not easily measured; but the deposit on the dorsal side of shells more than 8 years old can be easily flaked off and its thickness determined. The results are shown in Fig. 1. It will be seen that, exceptionally, a deposit 1·27 mm. thick can be laid down in 12 years, but most fall in the range 0·30–0·70 mm. in 17 years (1 mm. in 25–60 years). It is possible that some of the deposit may be rubbed off by the movements of the *Nucula*, although the umbo, which is always affected, probably projects above the surface of the mud and so avoids friction with the substratum. Maximum

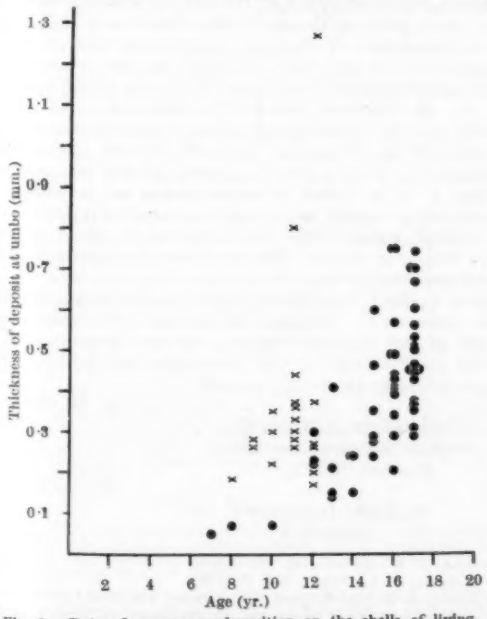

Fig. 1. Rate of manganese deposition on the shells of living *Nucula nucleus* (×) and *N. sulcata* (●)

Table 1

Locality	Manganese (per cent)
Nodules from Minard Narrows (Clyde)	18·7
Nodules from other sources (Goldberg) (ref. 3)	10·8–24·15
Substratum Arran Deep (Clyde) 60 fm.	0·1
Substratum North Sea 30 fm.	0·1
N. sulcata, Cumbrae Deep (Clyde), 60 fm.	8·5
N. sulcata, Arran Deep (Clyde), 60 fm.	17·9
Astarte montagui, Inverkip Bay (Clyde), 25 fm.	11·5

deposition of manganese occurs on shells and non-living loci at the surface of the bottom sediment[9]. The deposit on living molluscs is remarkably compact and hard, perhaps suggesting origin from biological activity rather than the adsorption of precipitated particles. The possible origin of the Clyde nodules and deposits from streams flowing off manganese and iron-rich bogs cannot be neglected, although a higher percentage of manganese in the bottom sediments might be expected. Such an origin was discussed by Murray and Irvine[9]. The figures given above suggest that the largest nodules (approx. 6 mm. diam.) in a sample taken from the Minard Narrows (L. Fyne) are 75–190 years old. This is in contrast with 3,000 years for deep-sea nodules of the same size.

I am indebted to Dr. A. Gilchrist of the Metallurgy Department, King's College, Newcastle upon Tyne, who kindly carried out the manganese estimations.

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PSYCHOLOGY

Scoring of a 'Steadiness' Test

A TEST of 'steadiness' has been used in this Department as a class experiment, in order to illustrate the importance of the 'double blind' principle in the design of controlled clinical trials. The apparatus of Seashore and Adams[1] was chosen. Steadiness is measured as the ability to hold a metal stylus in position for a fixed period within holes of graduated diameters, drilled in a metal plate, without allowing the stylus to come into contact with the sides of the hole. An index of steadiness is obtained by multiplying the contact times for each hole by arbitrary factors related to the hole diameters, and summing the result.

The timer used in this experiment was constructed from 'Add-on' dekatron units (Ericsson Telephones, Ltd., Beeston, Nottingham). A time signal (100 p.p.s.) is fed into the counting device through the input selector. With the 'count' key fixed in the

reset position, the counter is set up for automatic starting and resetting through the socket of the terminal dekatron unit, the connexions being made by means of a 12-pin miniature Jones plug. The counter starts operating when the terminal *PLA*9 is earthed and is reset by removing the earthing temporarily from *PLA*10. In the 'start' circuit there is a Post Office 'Uniselector' driven by 40 V. d.c. pulses from a time clock. The contacts in the 'Uniselector' are connected in such a way that, after a 2·5-sec. interval, the 'start' circuit is completed for a period of 10 sec. The 'start' circuit also contains a high-speed relay, which is actuated by a 40-V. d.c. current, 'made' when the stylus touches the sides of the hole. The relay closes the 'start' circuit and the counter operates. When the stylus is not touching the edge of the hole, the relay contacts are open, and the counter stops. Readings are thus obtained directly (in units of 0·01 sec.) of the time in a 10-sec. test period during which the stylus is in contact with the hole.

Experiments were carried out on 58 students aged 19–32, 54 of whom were aged 23 or less. The test procedure was as follows. Students were divided into pairs, one acting as subject and the other working the apparatus. The controlled testing situation for the stationary position recommended by Seashore and Adams[1] was adopted, with the exception that the apparatus allowed each subject a 2·5-sec. interval to become accustomed to the operation of the timer. A test run consisted of four replicate trials, each comprising one reading apiece in the five holes the dimensions of which are given in Table 1,*a*. After a control run, the subject swallowed a tablet, which might contain either a stimulant drug or a placebo. An hour later a further test run was carried out. In the conditions of the experiment, a 'placebo effect' could be demonstrated, contact times being significantly decreased ($P < 0.001$) at the second test run. No learning or fatigue effects during either run could be demonstrated.

Table 1. Correction Factors for a 'Stylus and Hole' Steadiness Test

Results of 232 observations on 58 subjects. To obtain a standardized score the contact times for each hole are multiplied by the appropriate correction factor and the results are summed

(*a*) Hole diameter (1/64 in.)	12	11	10	9	8
(*b*) Mean contact time in a 10 sec. test period (0·01 sec)	224·9	97·7	46·0	29·2	20·8
(*c*) $1/(b) \times 100$	0·445	1·023	2·174	3·425	4·808
(*d*) Correction factors (Spaeth and Dunham, ref. 2)	0·5	1	5	10	20
(*e*) Correction factors based on (*c*)	1	2	4	7	10

Mean contact times for each of the five holes were calculated from the control runs, comprising 232 readings in each hole (see Table 1,*c*). Some arbitrary assumption is necessary to render comparable the five respective means. At first sight it would appear simplest to use multiplication factors proportional to the reciprocals of the observed mean contact times so that, after correction, the readings from each hole would give, on the average, the same numerical result. The correction factors recommended by Spaeth and Dunham[2] (Table 1,*d*) do not obey this relation. An alternative series of integer factors which possesses approximately the desired property is given in Table 1,*e*. It is possible, however, that investigation of the wave form of upper limb tremor in relation to the geometry of the apparatus might show that some other basis for correcting the reading was preferable.

I wish to thank the students in the Pharmacology and Statistics Departments of the University of Sheffield who took part in the experiments, Mrs. W. M. Wright for help and statistical analyses and Dr. D. R. Wood for advice in constructing the apparatus.

J. D. Acland

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[1] Seashore, R. H., and Adams, R. D., *Science*, **78**, 285 (1933).
[2] Spaeth, R. A., and Dunham, G. C., *Amer. J. Physiol.*, **59**, 467 (1922).

STATISTICS

Fitting ν Linear Relations to n Variables all liable to Error

In a recent communication, B. E. Kelly[1] has directed attention to W. E. Deming's sadly neglected monograph "Statistical Adjustment of Data" (1943), and comments that Deming's procedure leads to the solution found by P. A. Wayman[2] for the best-fitting straight line to a series of observations of two variables both subject to variable error. This is not correct: Deming neglected the variation of his weighting factor W_i with the slope of the fitted line, and his result (p. 180) is a weighted regression. We have re-examined the problem of multivariate linear relations: when both variables are subject to independently variable errors we obtain a result the bivariate case of which agrees with Wayman's (which so far as we can find had not previously been described); when the ratio of the standard deviations of error in the several variables is constant for all sets of measurements, we find that for n variables connected by ν linear relations the best-fitting $(n - \nu)$-flat is defined by the ν latent vectors of the covariance matrix corresponding to its ν smallest latent roots; and we have been able to find a procedure suitable when not all the variables were measured at every point (a situation very common in studies of the correlation of chemical composition and physical properties of minerals). Certain non-linear relations of mineralogical importance are under investigation.

D. E. Barton and C. L. Mallows[3], discussing methods of curve-fitting other than least-squares, mention J. Berkson's method[4] of the 'controlled variable'. This method is based on the assumption that if in a series of experiments we adjust one variable as nearly as we can to a series of pre-selected 'control' values (one measurement at each) it will be proper to assume that the error of measurement is independent of the observed value, but that if no such 'control' is attempted the error of measurement is, necessarily, independent of the true value and not of the observed value; we are unable to find any justification for this postulate, and believe this procedure to be wholly invalid.

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[1] Kelly, B. E., *Nature*, **184**, 1086 (1959).
[2] Wayman, P. A., *Nature*, **184**, 77 (1959).
[3] Barton, D. E., and Mallows, C. L., *Nature*, **184**, 1086 (1959).
[4] Berkson, J., *J. Amer. Statist. Soc.*, **45**, 164 (1950). Cf. Lindley, D. V., *Biometrika*, **40**, 47 (1953).

FORTHCOMING EVENTS

*(Meetings marked with an asterisk * are open to the public)*

Monday, February 1

SOCIETY OF ENGINEERS (at the Geological Society, Burlington House, Piccadilly, London, W.1), at 5.30 p.m.—Mr. E. G. Massey Collier: Presidential Address.

SOCIETY OF CHEMICAL INDUSTRY, LONDON SECTION (at 14 Belgrave Square, London, S.W.1), at 6.30 p.m.—Mr. K. D. B. Johnson: "Industrial Separation of Isotopes".

Tuesday, February 2

PLASTICS INSTITUTE (at the Wellcome Building, 183–193 Euston Road, London, N.W.1), at 4 p.m.—Symposium on "Preplasticizing in Injection Moulding".

INSTITUTION OF ELECTRICAL ENGINEERS, MEASUREMENT AND CONTROL SECTION (at Savoy Place, London, W.C.2), at 5.30 p.m.—Dr. P. Vigoureux: "Development of the Formulæ of Electromagnetism in the M.K.S. System".

UNIVERSITY OF LONDON (at Imperial College of Science and Technology, South Kensington, London, S.W.7), at 5.30 p.m.—Prof. F. C. Tompkins: "Superficial Chemistry and Solid Imperfections".* (Inaugural Lecture.)

UNIVERSITY OF LONDON (at the London School of Hygiene and Tropical Medicine, Keppel Street, Gower Street, London, W.C.1), at 5.30 p.m.—Dr. Paul Fourman: "Magnesium Metabolism in Man".* (Seventh of fifteen lectures on "The Scientific Basis of Medicine" organized by the British Postgraduate Medical Federation. Further lectures on February 4, 9, 11, 16, 18, 23, 25 and March 1).

UNIVERSITY OF LONDON (at the Middlesex Hospital Medical School Mortimer Street, London, W.1), at 5.30 p.m.—Dr. Honor B. Fell: "The Experimental Transformation of Cells"*.

BRITISH INTERPLANETARY SOCIETY (at Caxton Hall, Westminster, London, S.W.1), at 7 p.m.—Mr. A. Wilson: "The Use of Reinforced Plastics in Rockets".

ROYAL AERONAUTICAL SOCIETY (at 4 Hamilton Place, London, W.1), at 7 p.m.—Mr. H. H. Pearcey: "Suppression of Shock-Induced Separations by Boundary Layer Control".

Wednesday, February 3

ROYAL SOCIETY OF ARTS (at John Adam Street, Adelphi, London, W.C.2), at 2.30 p.m.—Mr. Clifford Paine: "Modern Dyes" (Sir William Jackson Pope Memorial Lecture).

ROYAL STATISTICAL SOCIETY (at the London School of Hygiene and Tropical Medicine, Keppel Street, Gower Street, London, W.C.1), at 5 p.m.—Mr. R. L. Plackett: "Models in the Analysis of Variance".

SOCIETY OF CHEMICAL INDUSTRY, HEAVY ORGANIC CHEMICALS GROUP (at 14 Belgrave Square, London, S.W.1), at 6 p.m.—Mr. D. J. Hadley: "The Preparation and Use of Nitriles".

SOCIETY FOR ANALYTICAL CHEMISTRY (joint meeting with the OILS AND FATS GROUP of the SOCIETY OF CHEMICAL INDUSTRY, at the Chemical Society, Burlington House, Piccadilly, London, W.1), at 7 p.m.—Meeting on "Spectroscopic Investigation of Fats".

Thursday, February 4

ROYAL SOCIETY (at Burlington House, Piccadilly, London, W.1), at 10.30 a.m.—Discussion on "The Results of the Royal Society I.G.Y. Expedition" opened by Sir Graham Sutton, F.R.S.

PHYSICAL SOCIETY, OPTICAL GROUP (at Imperial College, London, S.W.7), at 2.30 p.m.—Dr. H. C. King: "The London Planetarium"; Mr. G. M. Sisson: "Present Plans for the Isaac Newton Telescope".

INSTITUTION OF NAVAL ARCHITECTS (at 10 Upper Belgrave Street, London, S.W.1), at 4.45 p.m.—Scientific Papers.

UNIVERSITY COLLEGE (in the Physiology Theatre, Gower Street, London, W.C.1), at 5 p.m.—Dr. Elizabeth Work: "The Mucopeptides of Bacterial Cell Walls".* (First of two lectures in Biochemistry. Further lecture on February 11.)

INSTITUTE OF REFRIGERATION (at the Institute of Marine Engineers, The Memorial Building, 76 Mark Lane, London, E.C.3), at 5.30 p.m.—Mr. J. M. Buist and Mr. R. Hurd: "Polyurethane Foams—Their Contribution to Heat Insulation".

INSTITUTION OF ELECTRICAL ENGINEERS (at Savoy Place, London, W.C.2), at 5.30 p.m.—Dr. J. H. Gridley "The Shielding of Overhead Lines against Lightning".

UNIVERSITY OF LONDON (at the London School of Hygiene and Tropical Medicine, Keppel Street, Gower Street, London, W.C.1), at 5.30 p.m.—Prof. E. B. Verney: "Some Aspects of the Renal Excretion of Water and Electrolytes".* (Eighth of fifteen lectures on "The Scientific Basis of Medicine" organized by the British Postgraduate Medical Federation. Further lectures on February 9, 11, 16, 18, 23, 25 and March 1.)

Friday, February 5

INSTITUTION OF ELECTRICAL ENGINEERS, MEDICAL ELECTRONICS DISCUSSION GROUP (at Savoy Place, London, W.C.2), at 5.30 p.m.—Discussion on "Computers in Medical Use" opened by Dr. R. A. Buckingham and Dr. J. M. Tanner.

SOCIETY OF DYERS AND COLOURISTS (at the Royal Society, Burlington House, Piccadilly, London, W.1), at 6 p.m.—Mr. L. Cotton and Mr. J. Wardle: "Dyeing and Finishing of 'Courtelon' Nylon in Relation to its Yarn Properties".

ROYAL INSTITUTION (at 21 Albemarle Street, London, W.1), at 9 p.m.—Prof. J. F. Danielli, F.R.S.: "Some New Approaches to Inheritance and Evolution" (The Woodhull Lecture).

Saturday, February 6

LONDON COUNTY COUNCIL (at the Horniman Museum, London Road, Forest Hill, London, S.E.23), at 3.30 p.m.—Miss Maitland Howard: "Cats and Magic".*

APPOINTMENTS VACANT

APPLICATIONS are invited for the following appointments on or before the dates mentioned:

ASSISTANT LECTURER IN PURE MATHEMATICS, and an ASSISTANT LECTURER IN APPLIED MATHEMATICS in the Department of Mathematics; and an ASSISTANT LECTURER IN APPLIED MATHEMATICS in the Department of Theoretical Mechanics—The Registrar, The University, Nottingham (February 6).

CHEMIST AND BACTERIOLOGIST (with an honours degree in chemistry, member of the Royal Institute of Chemistry, and extensive experience of water treatment and examination)—Fylde Water Board, Sefton Street, Blackpool (February 8).

LIBRARIAN (with a degree or recognized qualification in librarianship, experience in a special library, preferably serving a research organization), to take charge of the Library at the Central Electricity Generating Board's Research Laboratories at Leatherhead, Surrey—The Personnel Officer, Central Electricity Generating Board, 24/30 Holborn, London, E.C.1, marking envelope "Confidential Ref. N/9" (February 8).

ASSISTANT LECTURER IN BRITISH ARCHÆOLOGY—The Registrar, The University, Leicester (February 12).

ASSISTANT LECTURERS IN CHEMISTRY, PHYSICS and ZOOLOGY—The Registrar, The University, Leicester (February 12).

ASSISTANT LIBRARIAN (man or woman graduate)—The Registrar, The University, Leicester (February 12).

LECTURER or ASSISTANT LECTURER (with a good honours degree and experience in teaching and research) IN MATHEMATICS at the University of Malaya, Kuala Lumpur—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (London and Malaya, February 12).

PRINCIPAL OF CORNWALL TECHNICAL COLLEGE, Camborne—The Secretary for Education, County Hall, Truro, Cornwall (February 12).

LECTURER IN EDUCATION PSYCHOLOGY—The Registrar, The University, Liverpool, quoting Ref. CV/T (February 13).

RESEARCH ASSISTANT IN THE POSTGRADUATE DEPARTMENT OF APPLIED MECHANICS—The Registrar, The University, Sheffield (February 13).

RESEARCH ASSISTANT (with a good degree, and preferably a higher degree in physics, chemistry, engineering or ceramics) IN THE HOULDSWORTH SCHOOL OF APPLIED SCIENCE, to work on the high-temperature properties of certain ceramics of potential interest as engineering materials—The Registrar, The University, Leeds 2 (February 13).

GULBENKIAN RESEARCH FELLOW, to investigate certain aspects of the teaching of physics or chemistry in schools—The Registrar and Secretary, The University, Bristol (February 15).

LECTURER or DEMONSTRATOR IN CHEMICAL PATHOLOGY, for duties divided equally between original investigation and supervision of the hospital laboratory—The Registrar, The University, Leeds 2 (February 15).

READER IN APPLIED MATHEMATICS—The Clerk to the Governors, Chelsea College of Science and Technology, Manresa Road, London, S.W.3 (February 15).

EDITORIAL ASSISTANT, ASSISTANT EXPERIMENTAL or EXPERIMENTAL OFFICER, in the Publication Section, Road Research Laboratory, D.S.I.R., Harmondsworth, Middlesex, for work involving the editing and preparing for publication articles to technical journals, official booklets issued by the Laboratory, etc.—The Ministry of Labour, Technical and Scientific Register (X), 26 King Street, London, S.W.1, quoting A.21/0A (February 19).

JUNIOR LECTURER (preferably with the ability to share in the teaching of basic courses in psychology) IN EDUCATION—The Registrar, Trinity College, Dublin (February 22).

CHAIR OF ZOOLOGY at Westfield College—The Academic Registrar, University of London, Senate House, London, W.C.1 (February 25).

LECTURER IN PETROLOGY IN THE DEPARTMENT OF GEOLOGY—The Secretary, The University, Edinburgh (February 27).

TURNER AND NEWALL RESEARCH FELLOWS IN ENGINEERING, INORGANIC CHEMISTRY, PHYSICS and allied subjects—The Registrar, The University, Manchester 13 (February 27).

ASSISTANT LECTURER or LECTURER IN ANIMAL HUSBANDRY in the Faculty of Veterinary Science—The Registrar, The University, Liverpool, quoting Ref. CV/T (February 29).

LEVERHULME POSTGRADUATE FELLOWS (3), 2 IN THE FACULTY OF SCIENCE and 1 IN THE FACULTY OF ENGINEERING—The Registrar, The University, Liverpool, quoting Ref. CV/T (March 1).

CHAIR OF BIOCHEMISTRY at King's College, Strand, London, W.C.2—The Academic Registrar, University of London, Senate House, London, W.C.1 (March 2).

LECTURER or ASSISTANT LECTURER IN BOTANY at the University College of the West Indies—The Secretary, Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1 (March 5).

LECTURER or ASSISTANT LECTURER IN PHYSICS—The Registrar, The University, Southampton (March 5).

IMPERIAL CHEMICAL INDUSTRIES RESEARCH FELLOWS IN CHEMISTRY OR PHYSICS—The Registrar, The University, Hull (March 7).

ASSOCIATE PROFESSOR OF MATHEMATICAL STATISTICS at the University of New England, Australia—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Australia and London, March 12).

IMPERIAL CHEMICAL INDUSTRIES RESEARCH FELLOWS IN BIOCHEMISTRY, CHEMISTRY, ENGINEERING, PHARMACOLOGY or PHYSICS—The Secretary, The University, Aberdeen (March 12).

READER or SENIOR LECTURER IN AGRICULTURAL CHEMISTRY, and a LECTURER IN ANIMAL PHYSIOLOGY in the Faculty of Agriculture, Makerere College, University College of East Africa—The Secretary, Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1 (March 15).

SENIOR LECTURER (graduate of a British university or equivalent qualifications and experience appropriate to the post) IN PHYSICS; and an ASSISTANT LECTURER (with a good honours degree in physics and preferably some experience of teaching and research) IN PHYSICS at the University of Hong Kong—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Hong Kong and London, March 15).

LECTURER (preferably with a special interest in the field of transition elements or of complexes) IN INORGANIC CHEMISTRY—The Secretary, The Queen's University, Belfast (March 18).

LECTURER and an ASSISTANT LECTURER (preferably with a research interest in the application of mathematics to some engineering subject) IN ENGINEERING MATHEMATICS—The Secretary, The Queen's University, Belfast (March 30).

ISAAC WOLFSON RESEARCH FELLOW (suitably qualified graduate aged 24–35, either British subject (from any Commonwealth country) or Israeli citizen) IN METALLURGY—The Principal, St. Edmund Hall, Oxford (April 23).

ASSOCIATED ELECTRICAL INDUSTRIES FELLOW FOR RESEARCH IN THE DEPARTMENT OF PHYSICS—The Secretary-General of the Faculties, The Old Schools, The University, Cambridge (April 30).

ASSISTANT LECTURER IN THE DEPARTMENT OF NUTRITION—The Secretary, Queen Elizabeth College (University of London), Campden Hill Road, London, W.8.

ASSOCIATE DEAN OF THE SCHOOL OF ART AND SCIENCES; PROFESSOR OF ELECTRICAL ENGINEERING; PROFESSOR OF ENGINEERING PHYSICS; and a PROFESSOR OF MECHANICAL ENGINEERING, at the Middle East Technical University, Ankara—Room 139, Foreign Office, London, S.W.1.

CHEMIST (with a good honours degree in agricultural chemistry or chemistry and a knowledge of field experimentation) IN THE DEPARTMENT OF AGRICULTURE, Uganda, for research on soil fertility problems connected with tropical crop rotations, manures and fertilizers—The Director of Recruitment, Colonial Office, London, S.W.1, quoting BCD.63/9/09/T.

LECTURER (well qualified physicist, with research, industrial or teaching experience) IN THE SCIENCE AND MATHEMATICS DEPARTMENT—The Principal, Rugby College of Engineering Technology, Eastlands, Rugby.

LECTURER (with a degree in chemical engineering or metallurgy) IN THE DEPARTMENT OF METALLURGY, to teach the engineering principles of extraction metallurgy, particular attention being given to the effects of heat and mass transfer and fluid flow on the design and operation of metallurgical processes—Prof. F. D. Richardson, Department of Metallurgy, Imperial College of Science and Technology, Prince Consort Road, London, S.W.7.

LECTURER (with an honours degree in chemistry, with postgraduate experience of academic or industrial research) IN ORGANIC CHEMISTRY—The Secretary, Sir John Cass College, Jewry Street, London, E.C.3.

MASTER TO TEACH PHYSICS in the Upper School, including Sixth Forms—The Headmaster, Ellesmere College, Shropshire.

PHYSICIST, Senior grade (with at least five years experience in radiotherapy physics)—The Group Secretary, Mount Vernon Hospital, Northwood, Middlesex.

RESEARCH ASSISTANT (preferably with an honours degree in zoology or medical sciences, and a knowledge of German) IN THE DEPARTMENT OF ANATOMY, for work mainly on the central nervous system—The Secretary, University College, Gower Street, London, W.C.1.

RESEARCH ENTOMOLOGIST (male, with a good honours degree in natural science, or a degree in agriculture with honours in entomology, and experienced in entomological research) IN THE DEPARTMENT OF AGRICULTURE, Nyasaland, to assist with investigation of cotton pest problems in the Federation of Rhodesia and Nyasaland—The Director of Recruitment, Colonial Office, London, S.W.1, quoting BCD./63/4/016/T.

RESEARCH FELLOW (with a good degree in electrical engineering, physics or chemistry and some research experience of vacuum systems) IN THE DEPARTMENT OF ELECTRICAL ENGINEERING—The Registrar, Queen Mary College (University of London), Mile End Road, London, E.1.

SCIENTIFIC ASSISTANT, to assist an entomologist developing a technique to control *Culicoides impunctatus* in Scotland—R. Parish, B.Sc., Department of Zoology, University of Edinburgh, West Mains Road, Edinburgh 9.

REPORTS and other PUBLICATIONS

(not included in the monthly Books Supplement)

Great Britain and Ireland

Manchester Literary and Philosophical Society. Memoirs and Proceedings, Vol. 101, 1958–1959. Pp. 121+xii. (Manchester: Manchester Literary and Philosophical Society, 1959.) 31*s.* 6*d.* [312

De Havilland Propellers Limited. The De Havilland Transistor Curve Tracer. Pp. 4. Publication 1076: The De Havilland Portable Automatic Multi-core Cable Tester. Pp. 6. General Data. Pp. 7. Publication 5078: De Havilland Electromechanical Torque Motors—General Information. Pp. 8. (Hatfield: De Havilland Propellers Limited, 1959.) [312

Imperial College of Science and Technology. Inaugural Lectures, 1956–57 and 1957–58. Pp. ii+134+24 plates. (London: Imperial College of Science and Technology, 1959.) [312

Department of Scientific and Industrial Research and Fire Offices' Committee. Flame-Retardant Building Materials. Pp. iv+8. (London: H.M. Stationery Office, 1959.) 1*s.* net. [312

Privy Council. Medical Research Council Memorandum No. 38: Effect of Small Doses of Alcohol on a Skill Resembling Driving. By G. C. Drew, Dr. W. P. Colquhoun and Hazel A. Long. Pp. vi+108. (London: H.M. Stationery Office, 1959.) 7*s.* 6*d.* net. 312

British Museum (Natural History). Instructions for Collectors No. 11: Fossils, Minerals, and Rocks. Seventh edition. Pp. 16. 1*s.* Bulletin of the British Museum (Natural History). Historical Series. Vol. 2, No. 1: Darwin's Journal. Edited by Sir Gavin de Beer. Pp. 1–21. 8*s.* Entomology. Vol. 8, No. 5: The Walker Types of Fruit Flies (Tephritidae-Diptera) in the British Museum Collection. By D. Elmo Hardy. Pp. 159–242+plates 11–16. 30*s.* The John Murray Expedition, 1933–34. Scientific Reports. Vol. 10, No. 5: Sponges. By Dr. Maurice Burton. Pp. 151–281. 40*s.* Vol. 10, No. 6: Report on the Brachiopoda of the John Murray Expedition. By Dr. Helen M. Muir-Wood. Pp. 283–317+5 plates. 20*s.* (London: British Museum (Natural History), 1959.) [312

Other Countries

Optical Society of America, Washington. *Optics and Spectroscopy*. Vol. 6, No. 1. Translation of the Russian journal *Optika i Spektroscopiya*. Pp. iv+84. Published monthly. Subscription rate: 25 dollars plus 3 dollars for postage. (This rate includes the Journal of the Optical Society of America.) (New York: American Institute of Physics, 335 East 45th Street, 1959.) [3011

University of Natal. Wattle Research Institute (South African Wattle Growers' Union—University of Natal—Union Department of Forestry.) Report for 1958–1959 (Twelfth Year). Pp. 51+3 plates. (Pietermaritzburg: University of Natal—Wattle Research Institute, 1959.) [3011

Sitzungsberichte der Physikalisch-medizinischen Sozietät zu Erlangen, 79 Band, 1958. Herausgegeben im Auftrag der Sozietät von Hans-Jürgen Stammer. Pp. xviii+210. (Erlangen: Physikalisch-medizinische Sozietät, 1959.) [3011

Institut des Parcs Nationaux du Congo Belge. Exploration du Parc National de l'Upemba. Mission G. F. de Witte en collaboration avec W. Adam, A. Janssens, L. Van Meel et R. Verheyen (1948–1949). Fascicule 56: Amphibians. By Karl P. Schmidt and Robert F. Inger. Pp. 264+9 plates. (Bruxelles: Institut des Parcs Nationaux du Congo Belge, 1959.) [3011

Göteborgs Kungl. Vetenskaps-och Vitterhets-Samhälle. Report of the Swedish Deep-Sea Expedition, 1947–1948. Edited by Hans Pettersson. Vol. 6: Sediment Cores from the West Pacific. Pp. 99–160+14 plates. Sw. kr. 25. Vol. 10: Special Investigations. No. 4: On the Stratigraphy of some Bottom Sections from the Central Pacific. By F. Brotzen and A. Dinesen. Pp. 45–55. Sw. kr. 5. (Göteborg: Elanders Boktryckeri Aktiebolag, 1959.) [3011

Carnegie Institution of Washington. Publication No. 617: Miocene Floras of the Columbia Plateau. Part 1: Composition and Interpretation. By Ralph W. Chaney. Part 2: Systematic Considerations. By Ralph W. Chaney and Daniel I. Axelrod. Pp. viii+237+44 plates. (Washington, D.C.: Carnegie Institution of Washington, 1959.) 4.25 dollars; cloth bound 4.75 dollars. [3011

International Atomic Energy Agency, Vienna. Assistance Through Exchange of Nuclear Scientists. Pp. 8. A Chronological List of Atomic Energy Conferences, Meetings, Training Courses (Issue No. 5). Pp. 22. (Vienna: International Atomic Energy Agency, 1959.) [312

World Health Organization. Bulletin. Vol. 21, No. 1: Epidemiological Basis of Tuberculosis Eradication in Denmark. A Concurrent Comparison of Home and Sanatorium Treatment of Pulmonary Tuberculosis in South India. Pp. v+144. (Geneva: World Health Organization; London: H.M. Stationery Office, 1959.) 6 Swiss francs; 10*s.*; 2 dollars. [312

World Health Organization. Technical Report Series, No. 181: Immunological and Hæmatological Surveys—Report of a Study Group. Pp. 35. 1 Swiss franc; 1*s.* 9*d.*; 0.30 dollars. Communicable Diseases in Schools: a Survey of Existing Legislation. Pp. 65. 2 Swiss francs; 3*s.* 6*d.*; 0.70 dollars. [312

Canada: Department of Mines and Technical Surveys. Geological Survey of Canada. Economic Geology Series No. 17: Tungsten Deposits of Canada. By H. W. Little. Pp. viii+251. 1.50 dollars. Memoir No. 288: Surficial Geology of the Lindsay-Peterborough Area, Ontario, Victoria, Peterborough, Durham, and Northumberland Counties, Ontario. By C. P. Gravenor. Pp. 60+7 plates. 1 dollar. (Ottawa: Queen's Printer, 1959.) [312

United States Department of the Interior: Geological Survey. Water-Supply Paper 1320-E: Summary of Floods in the United States during 1953. Prepared under the direction of J. V. B. Wells. Pp. v+341–364. 20 cents. Water-Supply Paper 1505: Surface Water Supply of the United States 1957. Part 3-A: Ohio River Basin except Cumberland and Tennessee River Basins. Prepared under the direction of J. V. B. Wells. Pp. xii+553. 1.75 dollars. Professional Paper 315-A: Geology of Part of the Horseshoe Atoll in Scurry and Kent Counties, Texas. By Philip T. Stafford. Pp. iii+20+8 plates. (Washington, D.C.: Government Printing Office, 1959.) [312

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